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## THREE ZOÖPAGACEOUS FUNGI THAT CAPTURE AND CONSUME SOIL- INHABITING RHIZOPODS

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(WITH 8 FIGURES)

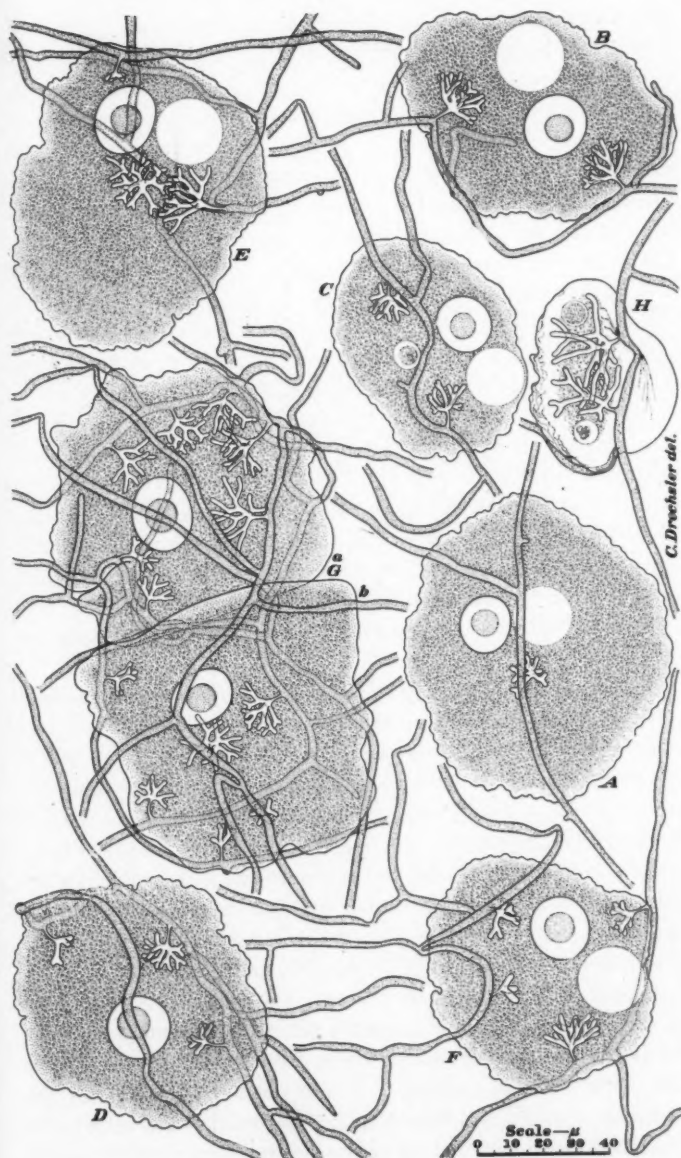
Three fungi, all subsisting by capture of terricolous rhizopods, are described herein as new species of the Zoöpagaceae. Two of the new forms can be set forth in their asexual and sexual reproductive phases as well as their vegetative stage; though the details of morphology whereby they differ from forms previously made known appear of rather commonplace character. Their prey, like the prey of most predaceous members of the family, and, indeed, like the host animals of most parasitic members, consists of amoebae of the familiar pelliculate type developing abundantly in agar plate cultures planted with partly decayed vegetable materials. Although the third form can be presented only in its vegetative and asexual reproductive stages, the vegetative stage here offers marked departure in its predaceous relationship to a non-pelliculate, frequently reticulate rhizopod, while the asexual reproductive phase is given distinctiveness by the unusual design of the curiously topknotted conidia.

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A RHABDOSPORA-LIKE SPECIES OF STYLOPAGE SUBSISTING ON  
AMOEBIA VERRUCOSA

A maize-meal-agar plate culture which, after being permeated with mycelium of *Pythium ultimum* Trow, had been further planted with a small quantity of partly decomposed friable barley (*Hordeum vulgare* L.) straw collected near Greeley, Colorado, early in October, 1945, showed after 20 days dozens of large amoebae congregated in areas adjacent to the superadded material. On microscopical examination under low magnification the clustered animals gave much the same appearance as had been noted earlier in the groups of large amoebae captured by my *Zoöpage phanera* (3: 26-30) and more recently again in the assemblages of robust individuals of *Amoeba terricola* Greeff (*sensu strictiore*) taken captive by my *Acaulopage marantica* (8: 143-149). Under higher magnification the captured animals (FIG. 1, A-F; G, a, b; H), which commonly measured 50 to 100  $\mu$  across, were each found to be surrounded by a clearly visible, firm pellicle, in part delicately rippled and in part disposed more smoothly about broadly protruding pseudopodia. In the colorless, rather dispersedly granular sarcode could readily be distinguished a single somewhat prolate ellipsoidal nucleus measuring commonly 17 to 23  $\mu$  in length and 14 to 19  $\mu$  in width. As the nucleus showed always a slightly darker globose central body, mostly 7.5 to 10.5  $\mu$  in diameter, within the clear outer layer, the animal was immediately recognized as *Amoeba verrucosa* Ehrenb.—as the same widespread soil-inhabiting rhizopod that previously has been found attacked endoparasitically by *Cochlonema megalosomum* Drechs. (8: 128-137), *C. symplocum* Drechs. (9: 258-266), and *C. agatum* Drechs. (12: 120-133). Although this rhizopod has further been reported as subject to destruction by the predaceous hyphomycete *Dactylella tylopaga* Drechs. (4), it has not hitherto been found captured by any member of the Zoöpagaceae.

In the areas where capture of *Amoeba verrucosa* was just beginning to become noticeable from a grouped arrangement of relatively few individuals, the animals were often held through adhesion to only a single meagerly branching mycelial filament (FIG. 1, A), mostly 1.4 to 3  $\mu$  wide, whose phycomycetous character was

FIG. 1. *Stylopage rhabdoides*.

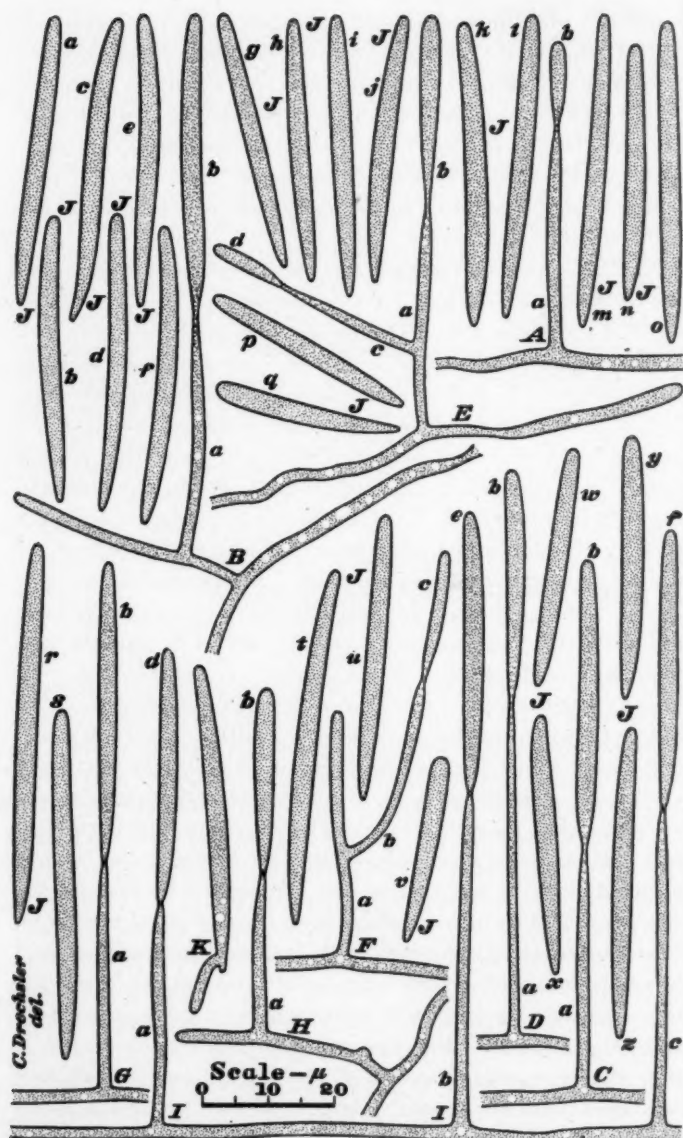
shown clearly in a lack of all intercalary cross-walls. After an animal had been taken, branches were evidently put forth more abundantly from the adhering portions of filament than from adjoining portions; so that the captive became increasingly invested with hyphal elements (FIG. 1, *B-D*). As the rhizopod usually continued long to wallow about by extending pseudopodia now in one direction now in another, it was not surprising that the investing hyphae were soon found to pursue markedly irregular courses. Yet even where the animal's struggles seemed less resolute the new hyphae that were put forth locally appeared likewise much given to capricious turns and to rather pronounced haphazard variations in width. Similar haphazard irregularity in the elongation of enveloping branches was observable also in instances where the rhizopod was captured by two (FIG. 1, *E, F*) or more (FIG. 1, *G, a, b*) separate hyphae, as often happened where the fungus had been present for some time and had thus been able beforehand to extend mycelial filaments more copiously. Consequently in areas where many captives had become unhappily congregated in readily noticeable groups, the animals, whether single or huddled in pairs (FIG. 1, *G, a, b*), were nearly always found intricately enveloped, above and below, in a confusion of promiscuously branching hyphae.

Usually at a rather early stage in the envelopment of the captured animal, a narrow process, or sometimes two narrow processes, would be extended through its pellicle from the adhering filament or filaments. On attaining a length of several microns each of the processes would widen abruptly at the tip and then would branch dichotomously at close intervals to form a pedicellate haustorium with short divaricate assimilative branches. Further envelopment of large animals usually brought intrusion of additional haustoria, so that in the end six or seven such organs were commonly found present (FIG. 1, *G, a, b*), and in more than a few instances as many as ten or twelve. The progressive expropriation of materials by these haustoria did not immediately have any noticeable effect on the sarcoderm, or on the nucleus, or on the operation of the contractile vacuole. It was not until the animal's protoplasmic contents had been largely depleted that the nucleus degenerated visibly and that the contractile vacuole ceased to operate.



Sometimes when the last remnants of degenerating protoplasm lay beyond reach of the haustoria many of the assimilative branches most favorably situated would elongate at a relatively late stage (FIG. 1, *H*) to bring about thorough expropriation. In any case, when all granular residues had vanished, the contents of the haustoria themselves were withdrawn backward into the parent mycelium, leaving only the collapsed empty pellicle as evidence of the completed predaceous action.

Near the groups of captured animals the Colorado fungus gave rise on prostrate hyphae to erect conidiophores (FIG. 2, *A-E*: *a*) which at a height of 25 to 50  $\mu$  tapered rather markedly before they widened again in elongating further to form the single terminal conidium (FIG. 2, *A-E*: *b*). Sometimes the conidiophore (FIG. 2, *E*, *a*) growing out distally into a young conidium (FIG. 2, *E*, *b*) extended a lateral branch (FIG. 2, *E*, *c*) which soon began to form distally a second young conidium (FIG. 2, *E*, *d*). Occasionally the erect hypha (FIG. 2, *F*, *a*) would fail to produce a conidium at its tip, but instead gave rise to a lateral branch (FIG. 2, *F*, *b*) that subsequently was found producing a young conidium (FIG. 2, *F*, *c*). When definitive size was attained, two cross-walls were laid down close together in the narrow isthmus to separate each functional conidiophorous hyphal element (FIG. 2, *H*, *a*; *I*, *a-c*) from the conidium (FIG. 2, *H*, *b*; *I*, *d-f*). No instance of a conidiophore growing from below its delimited distal end to form a second conidium on a newly prolonged tip came to light in the fungus preying on *Amoeba verrucosa*. Absence of such successive development, and lesser length of the conidiophore—this dimension here did not usually exceed 50  $\mu$ —would seem to represent features distinguishing the fungus from my *Stylopage rhabdospora* (5: 374-377; 12: 138-140), in which production of plural conidia on a successively elongated conidiophore has often been observed, and in which the conidiophores, even without taking any increments into account, have frequently been found measuring between 50 and 100  $\mu$  in height. With respect to shape the conidia (FIG. 2, *J*, *a-s*) closely resemble those of *S. rhabdospora*, and though they have been found of appreciably larger size the dimensional difference is not sufficiently pronounced to merit much emphasis.

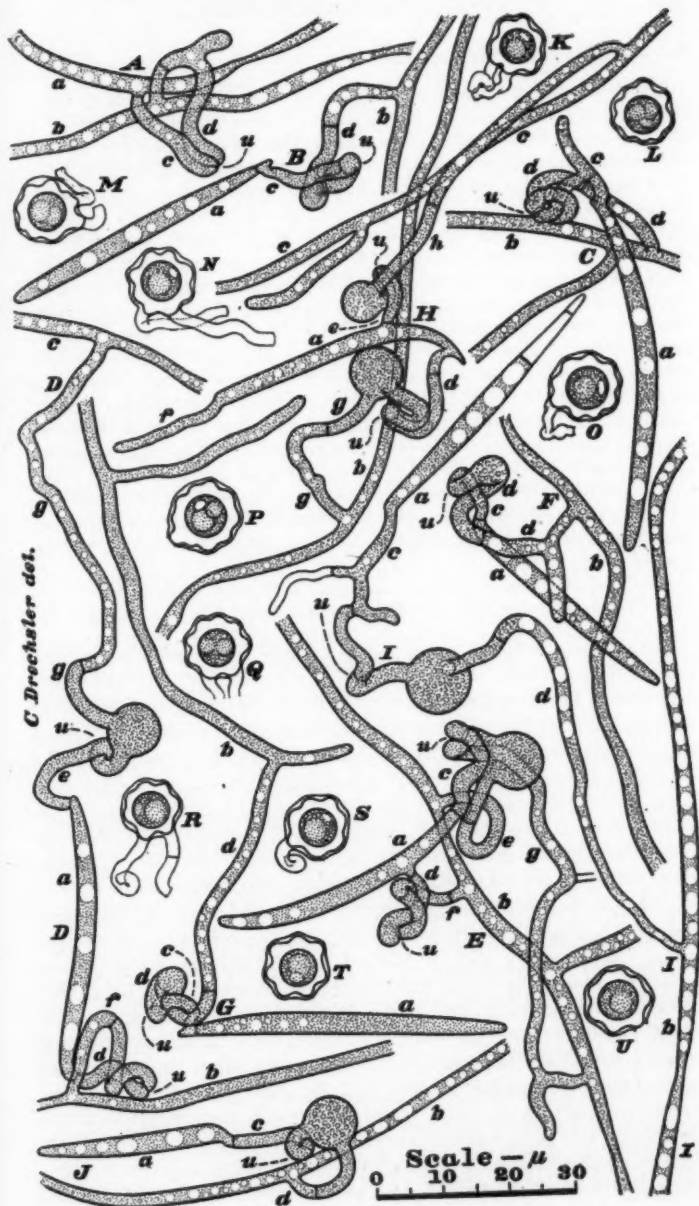
FIG. 2. *Stylopage rhabdoides*.

In nearly all areas occupied by it the fungus predaceous on *Amoeba verrucosa* showed moderately abundant sexual reproduction. Now and then instances were found where two neighboring mycelial filaments (FIG. 3, A, a, b) had evidently given rise to a pair of zygothoric branches (FIG. 3, A, c, d) which, after making contact with each other distally, became united at the tip (FIG. 3, A, u); a septum having meanwhile been laid down in each branch about 15 to 25  $\mu$  from the tip to delimit the conjugating parts as gametangia. Much more often, however, one of the two conjugating elements was supplied from a germinating conidium (FIG. 3, B-J: a) while the other was supplied from a mycelial hypha (FIG. 3, B-J: b; D, c; H, c). Most commonly the conidium germinated from one of its ends (FIG. 2, K). Sometimes the cross-wall delimiting the gametangium was laid down within the conidium itself, so that a small portion of the asexual spore was included in the conjugating cell (FIG. 3, B, c; E, c; G, c). In other instances the delimiting wall of the gametangium was laid down at the base of the germ-tube flush with the conidial wall (FIG. 3, D, d, e; F, c; H, e). Frequently, again, the septum was intercalated in the germ-tube, whether within a few microns from the base (FIG. 3, C, c; E, d; H, d; J, c) or, perhaps, 10 to 25  $\mu$  above it (FIG. 3, I, c). Although the opposing gametangium borne on a branch from a mycelial hypha sometimes included nearly the entire branch (FIG. 3, E, f), more often the delimiting septum here was found 5 to 25  $\mu$  above the branch origin (FIG. 3, B, d; C, d; D, f; F, d; H, g; J, d) and not infrequently it would set off the sexual cell from a stalk more than 25  $\mu$  (FIG. 3, E, e; G, d; H, h) or even more than 50  $\mu$  (FIG. 3, D, g; I, d) in length. Some conidia participated in the development of two zygosporos by giving rise to two germ tubes that each produced a gametangium. In such instances the two germ-tubes came from opposite ends of the spore (FIG. 3, D, d, e), or, again, one of the two came from an end position (FIG. 3, E, c; H, d) while the other came from a more nearly median position (FIG. 3, E, d; H, e). Often the two gametangia borne on the germ-tubes conjugated with gametangia from separate mycelial hyphae (FIG. 3, D, b, c; H, b, c), but rather frequently, also, they conjugated with gametangia supplied from the same mycelial filament (FIG. 3, E, b).

In one observed instance a gametangium (FIG. 3, E, c) supplied by a conidium became united with two sexual branches (FIG. 3, E, e, g) supplied from the same mycelial filament. Occasionally a conidium that had already supplied two gametangia (FIG. 3, H, d, e) on separate germ-tubes was found putting forth a third germ-tube (FIG. 3, H, f).

Following fusion of the paired gametangia one of them—more usually the one contributed from the mycelial filament—soon began to swell out, commonly at a distance of about  $5\mu$  from the union, to form a lateral intercalary globose excrescence (FIG. 3, D, g; E, e; H, g, h; I, d; J, d). When this excrescence attained a diameter of about  $10\mu$ , and had received the entire protoplasmic content of both gametangia, it gave rise endogenously to the zygosporer. At maturity the membrane of the globose part enveloped somewhat loosely the boldly verrucose, thick-walled, distinctly yellowish sexual spore (FIG. 3, K–U). Internally the ripe zygosporer often seemed to have the unitary organization familiar among oöspores, its coarsely granular material apparently surrounding a single reserve globule and single refringent body (FIG. 3, K, N, O, R, S, T, U). At other times, however, two or three homogeneous reserve globules seemed present, usually without any clearly discernible refringent body (FIG. 3, L, M, P, Q).

The fungus manifestly is most closely related to *Stylopage rhabdospora*. In the vegetative stage it appears separated from that species by reason of its coarser mycelial hyphae and its utilization of a different *Amoeba* as prey. Though not wholly unknown in the Zoöpagaceae, utilization of more than one species of rhizopod is so exceptional among members of the family that it cannot be assumed for any member without unmistakable evidence. In view of the differences shown in its vegetative stage and in its conidiophores, there seems somewhat more reason for holding the fungus to be distinct from *S. rhabdospora* than for regarding it as being the same. It is therefore described as new under a specific epithet meaning "rod-like," which may perhaps serve helpfully in signaling its conidial shape and in recalling the species most closely related to it.

FIG. 3. *Stylopage rhabdoides*.

**Stylopage rhabdoides** sp. nov.

Mycelium effusum; hyphis continuis, incoloratis, filiformibus, parce ramosis, fere  $1.4-3\ \mu$  crassis, ad animalia minuta extense inhaerentibus, saepe ea contorte implicantibus, pelliculam eorum perforantibus, haustoria intus evolvuntibus quae protoplasma exhauriunt; haustorio pedicellato, pedicello vulgo  $3-7\ \mu$  longo,  $0.5-1.5\ \mu$  crasso, apice abrupte latescente, saepius bis vel quater repete bifurco, ita usque 16 ramulos divaricatos  $5-18\ \mu$  longos et  $1-2.5\ \mu$  crassos ferente. Hyphae fertiles incoloratae, erectae, simplices vel interdum parce ramosae, vulgo  $20-50\ \mu$  altae,  $1.4-2\ \mu$  crassae, sursum leniter attenuatae, apice unicum conidium gignentes; conidiis incoloratis, elongato-cylindraceis, saepe sursum leniter attenuatis et abrupte rotundatis, deorsum plus attenuatis, itaque basi acutiusculis, plerumque  $25-57\ \mu$  longis,  $2.7-3.5\ \mu$  crassis. Hyphae zygosporiferae saepius  $15-85\ \mu$  longae,  $1.5-3\ \mu$  crassae, vulgo  $10-27\ \mu$  infra apicem septo divisa, ambae rarenter ex duabus hyphis mycelii exeuntes sed saepissime altera ex hypha mycelii altera ex conidio germinanti oriunda; zygospores cellulis terminalibus (gametangiis) saepe plus minusve irregulariter flexuosis, interdum inter se circumplicantibus, apice inter se conjungentibus; zygosporangio circa  $5\ \mu$  ab junctione oriundo, primum levi, sphaeroideo, vulgo  $9-11\ \mu$  crasso, maturitate membrana ejus circa zygosporam laxè collapsa; zygospora aliquantum flavida, globosa, circa  $8-10\ \mu$  crassa, in maturitate membrana valde verrucosa, cellulam viventem sphaeralem  $5-6.5\ \mu$  crassam circumdante.

Amoebam verrucosam capiens consumensque habitat in stramento (foliis acere caulibusque) *Hordei vulgaris* putrescenti prope Greeley, Colorado.

Mycelium spreading; vegetative hyphae colorless, filamentous, sparingly branched, mostly  $1.4-3\ \mu$  wide, adhering to and often extensively enwrapping minute animals, penetrating the pellicle of each captive and intruding haustoria to appropriate the protoplasmic contents; haustoria pedicellate, the pedicel usually  $3-7\ \mu$  long,  $0.5-1.5\ \mu$  wide, abruptly enlarging distally and bifurcating often 2 to 4 times in succession at wide angles, thus bearing commonly 4 to 16 assimilative branches  $1-2.5\ \mu$  wide and in combined length of successive parts measuring  $5-18\ \mu$ . Conidiophores simple or occasionally sparingly branched, colorless,  $20-50\ \mu$  high,  $1.4-2\ \mu$  wide, tapering gradually toward the apex whereon a single conidium is borne; conidia colorless, elongated-cylindrical, tapering slightly toward the abruptly rounded tip and rather pronouncedly toward the somewhat more pointed basal end, mostly  $25-57\ \mu$  long and  $2.7-3.5\ \mu$  wide. Zygosporic hyphae mostly  $15-85\ \mu$  long and  $1.5-3\ \mu$  wide, both occasionally rising from 2 mycelial filaments, but much more often one of a pair arising from a mycelial filament and the other from a germinating conidium, each in any case partitioning off a terminal cell  $10-27\ \mu$  long; the paired terminal cells often more or less irregularly flexuous and sometimes intertwined, conjugating apically. Zygosporangium formed about  $5\ \mu$  from the union and mostly in the

gametangium supplied from the mycelium, at first smoothly subspherical and commonly  $9-11\ \mu$  in diameter, its membrane at maturity collapsing loosely about the zygospore; the latter distinctly yellowish, subspherical, boldly verrucose,  $8-10\ \mu$  in diameter, its thick wall surrounding a living cell  $5-6.5\ \mu$  in diameter.

Capturing and consuming *Amoeba verrucosa* it occurs in decaying straw (stems, leaves, and chaff) of *Hordeum vulgare* near Greeley, Colorado.

A SLENDER-SPORED ACAULOPAGE PREYING ON TWO SPECIES  
OF AMOEBA

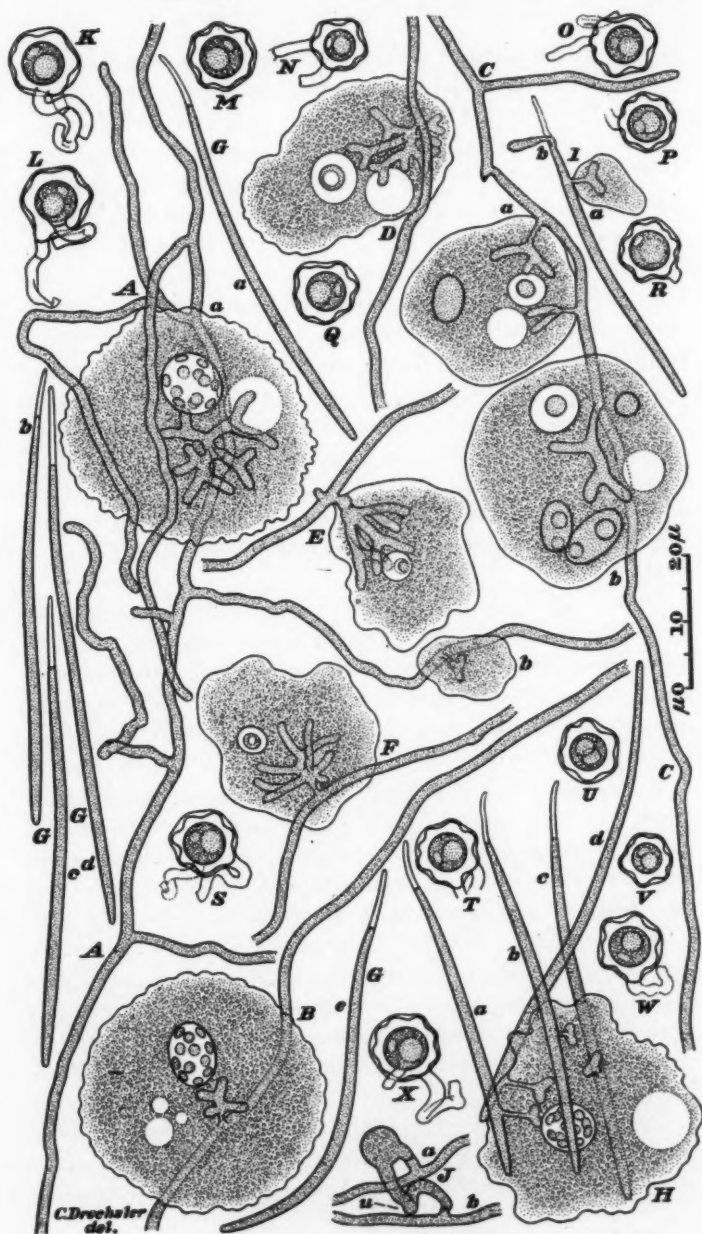
Several maize-meal-agar plate cultures which after being permeated with mycelium of *Pythium ultimum* had been further planted with small quantities of deciduous leaf mold collected near Mercer, Wisconsin, on November 14, 1945, showed in ten days a somewhat extensive development of an aseptate mycelium composed of hyphae about  $1.5\ \mu$  wide that were active in capturing amoebae belonging manifestly to two separate species. The captured animals referable to one species (FIG. 4, A, a; B) often measured about  $35\ \mu$  across when they were drawn into a rounded shape. The delicate and frequently rather minutely rippled pellicle here surrounded finely granular protoplasm together with a single globose or prolate ellipsoidal nucleus, commonly  $8$  to  $9.5\ \mu$  long and  $6$  to  $8\ \mu$  wide, in which about twelve slightly darker oblate ellipsoidal bodies could be distinguished in scattered positions close under the peripheral membrane. Unquestionably these captives were referable to the same species of *Amoeba* that previously had been found captured habitually by my *Zoöpage thamnospira* (7: 141-144) and my *Acaulopage tetraceros* (10: 289-291). The other animals serving as prey measured commonly  $30$  to  $35\ \mu$  across when drawn into a rounded form (FIG. 4, C, a, b; D; E; F). They similarly were surrounded by a thin firm pellicle, and their protoplasm, too, was of finely granular, hyaline character; but the single globose or prolate ellipsoidal nucleus they contained, which often measured  $4.3$  to  $8\ \mu$  in length and  $4$  to  $7\ \mu$  in width, had its darker material collected in a globose central body,  $2.2$  to  $3\ \mu$  in diameter, that frequently offered a homogeneous appearance (FIG. 4, C, a, b) but frequently, again,



revealed a vacuole or lacuna of variable size (FIG. 4, *D*, *F*). While for purposes of identification this type of nuclear organization is less distinctive than might be desired, occurring evidently among many species of *Amoeba*, large and small, it nevertheless separates decisively any animal that embodies it from animals whose darker nuclear material (chromatin) is present in plural peripheral bodies.

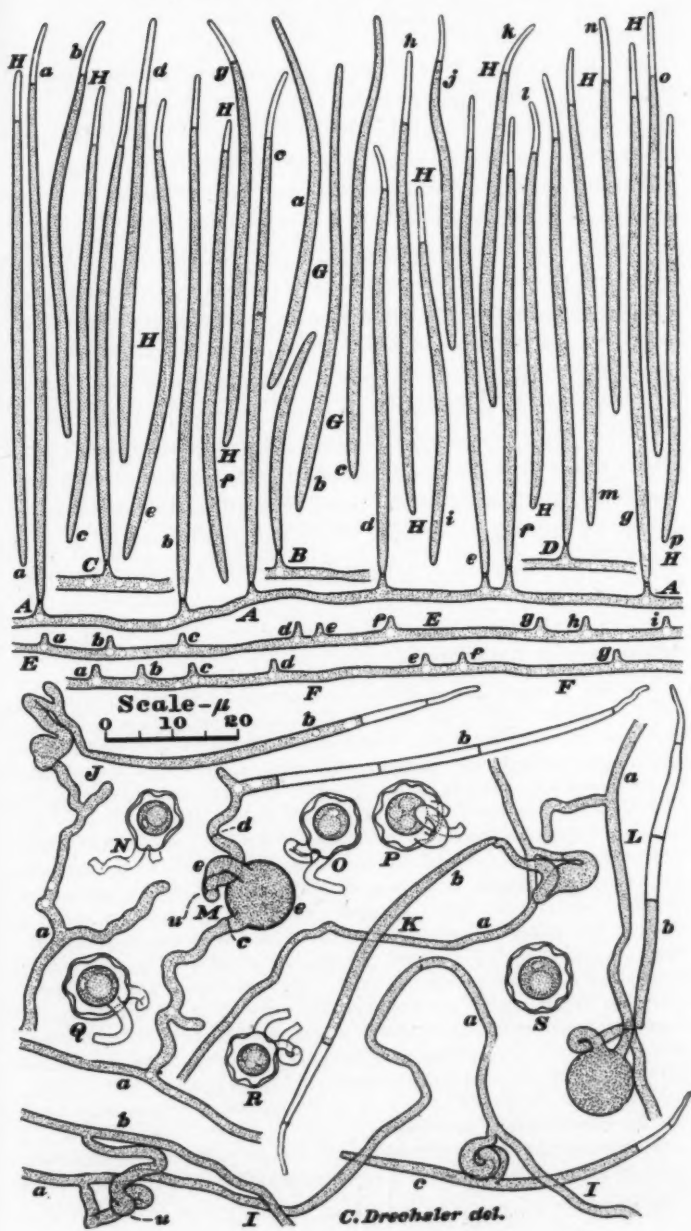
Individuals of both species of *Amoeba* were held securely through adhesion to the aseptate hyphae. Usually the captive was invaded by only a single haustorium (FIG. 4, *A*, *a*, *b*; *B*; *C*, *b*; *E*; *F*), though occasionally two haustoria were intruded (FIG. 4, *C*, *a*). The assimilative organs were of the pedicellate type, consisting of a usually slender pedicel together with divaricate absorptive branches approximately equal in width to the mycelial hyphae. In some instances the pedicel seemed wider than is usual for this structure (FIG. 4, *C*, *b*) and in others appeared, besides, somewhat shortened (FIG. 4, *E*). The captured animals of both species remained alive, stubbornly operating their contractile vacuoles until most of their protoplasmic material was expropriated. When ultimately death came, the nucleus degenerated; its degenerating substance and all granular remnants of cytoplasm then being assimilated by the fungus. Subsequently the contents of the haustorium were withdrawn backward into the parent hypha, leaving only the empty evanescent envelope of the absorptive apparatus within the equally evanescent pellicle of the rhizopod.

Thus amply nourished from abundant prey the fungus gave rise freely to asexual reproductive apparatus consisting of long, slender conidia (FIG. 5, *A*, *a-g*; *B-D*) borne erectly on short sterigmata projecting upward from procumbent hyphae (FIG. 5, *A-D*; *E*, *a-i*; *F*, *a-g*). In some instances a conidium was found delimited from its sterigma by a basal septum while protoplasm still filled the young spore throughout its length (FIG. 5, *B*). Conidia filled with protoplasm from base to tip were often found detached (FIG. 5, *G*, *a-c*) after being subjected to the disturbance unavoidable in covering material with a cover glass. In undisturbed cultures the conidia, after being delimited at the base, usually remained attached while a distal portion, including from one-tenth to one-fifth of the length of spore, was evacuated of contents; so that, as

FIG. 4. *Acaulopage ischnospora*.

a rule, spores that had become detached spontaneously bore an empty portion of tubular envelope as an apical appendage (FIG. 4, *G, a-e*; FIG. 5, *H, a-p*). Whether they bore an empty appendage (FIG. 4, *H, a-c*) or were filled with protoplasm throughout (FIG. 4, *H, d*), detached conidia were readily capable of infecting a susceptible amoeba by adhering to its pellicle and then invading its sarcode with a haustorium. Usually animals attacked in such commonplace parasitic manner, especially when they were of good size, continued in their locomotion for some time, carrying the spores along with them. However, when a conidium intruded a haustorium into a smaller adhering amoeba measuring perhaps only about  $10\mu$  across (FIG. 4, *I, a*)—a nucleus could not usually be distinguished in these undersized specimens—the animal's locomotion was arrested no less decisively than when capture was effected by a mycelial filament (FIG. 4, *A, b*). As might be expected, capture and invasion of an amoeba by a conidium did not prevent the conidium from putting forth a vegetative germ-tube (FIG. 4, *I, b*).

Usually after asexual reproduction had been proceeding for some time the fungus also produced sexual apparatus in readily noticeable quantity. As in *Stylopage rhabdoides* and many other zoöpagaceous forms, zygospore development took place only sparingly before conidia were formed; for here, too, zygophoric branches from mycelial hyphae (FIG. 4, *J, a, b*; FIG. 5, *I, a, b*) only occasionally would pair and conjugate with others of similar origin. Once a tract of substratum became bestrewn with detached conidia, however, more spirited conjugation ensued between sexual branches arising from mycelium filaments (FIG. 5, *I-M: a*) on the one hand, and sexual branches arising as germ-tubes from conidia (FIG. 5, *I, c*; *J-M: b*) on the other. Owing to the slenderness and frequently rather irregular course of the conjugating hyphal elements the cross-walls delimiting the gametangia were often not clearly discernible; and the place of union between the gametangia often remained uncertain for similar reasons. Where both delimiting septa (FIG. 5, *M, c, d*) and the apical union (FIG. 5, *M, u*) were clearly revealed, the globose enlargement destined for the formation of the zygospore was more often found developing in the gametangium on the branch contributed by the my-

FIG. 5. *Acanlopage ischnospora*.

celial hypha (FIG. 5, *M*, *e*) than in the gametangium borne on the germ hypha. As in many related species the progressive evacuation of the conidium entailed in the growth of the zygosporangium was marked by deposition of two to four successive retaining walls within the conidial envelope (FIG. 5, *J-M*: *b*). When the sexual apparatus reached maturity (FIG. 4, *K-W*; FIG. 5, *N-S*) the membrane of the globose zygosporangium was found collapsed somewhat loosely about a pronouncedly verrucose, thick-walled, yellowish zygosporangium that showed an internal organization rather similar to the unitary organization familiar in the oöspores of many oömycetes—their granular protoplasm being disposed in a parietal layer surrounding a homogeneous central reserve globule.

In the genus *Acaulopage* to which it manifestly belongs, the fungus would seem to be distinguished from previously described species more especially by the unusual length and slenderness of its distally appendaged conidia. It is therefore presented as new under a specific epithet compounded in part of a word meaning both "thin" and "withered."

***Acaulopage ischnospora* sp. nov.**

Mycelium effusum; hyphis continuis, incoloratis, filiformibus, parce ramosis, plerumque  $1-2\ \mu$  crassis, ad animalia minuta inhaerentibus, pelliculam eorum perforantibus, haustorium (quandoque 2 haustoria) intus evolventibus quod protoplasma exhaurit; haustorio pedicellato, pedicello fere  $2-6\ \mu$  longo,  $0.5-1\ \mu$  crasso, apice abrupte latescente, semel vel quater repetite bifurco, ita  $2-12$  ramulos divaricatos  $2-12\ \mu$  longos  $1-2\ \mu$  crassos ferente. Conidia incolorata, ex sterigmatibus erectis, plerumque  $1.5-4\ \mu$  altis, basi  $1-1.5\ \mu$  crassis, sursum attenuatis, apice  $0.5-0.7\ \mu$  crassis, inter se  $3-25\ \mu$  distantibus oriunda, vulgo in partibus duabus constantia: pars supera vacua,  $4-13\ \mu$  longa, basi  $0.8-1.2\ \mu$  crassa, sursum leniter attenuata, saepius plus minusve marcida vel collapsa; pars infera protoplasmatis repleta, filiformis, utroque parvulum attenuata, plerumque  $50-80\ \mu$  longa,  $1.6-2\ \mu$  crassa. Hyphae zygosporiferae irregulariter flexuosae, quandoque inter se circumplicantes, ambae rarerent ex duabus hyphis mycelii exeuntes sed saepissime altera ex hypha mycelii altera ex conidio germinanti oriunda. Zygosporangia primo levia, sphaeroidea, plerumque  $8-11\ \mu$  crassa, membrana eorum in maturitate circa zygosporam laxè collapsa; zygospora aliquantum flavida, globosa,  $7-10\ \mu$  crassa, valde verrucosa, membrana ejus  $1-2\ \mu$  crassa cellulam viventem sphaeralem  $4.3-6.5\ \mu$  crassam stricte circumdante.

*Amoebas* duarum specierum  $20-40\ \mu$  latas capiens consumensque habitat in foliis arborum (*Betulae*, *Aceris*, *Ulmi*) putrescentibus prope Mercer, Wisconsin.

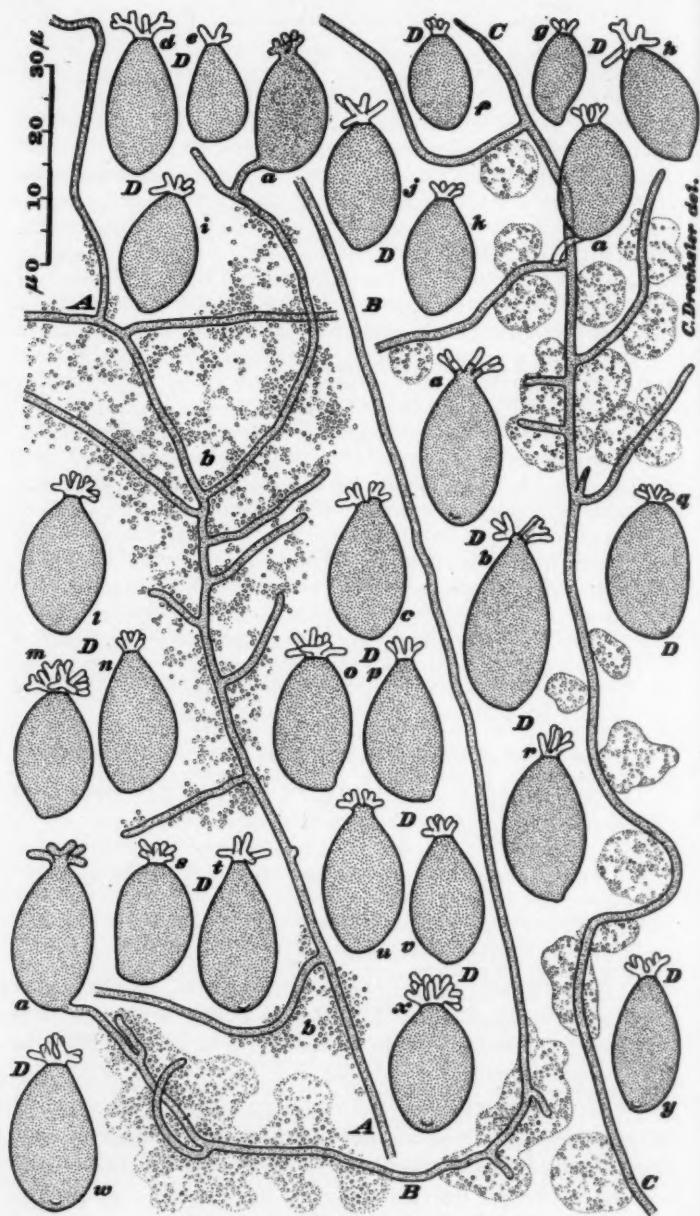
Mycelium spreading; vegetative hyphae continuous, colorless, filamentous, sparingly branched,  $1-2\ \mu$  (mostly about  $1.5\ \mu$ ) wide, adhering to minute animals, penetrating the pellicle of each animal thus captured, and intruding a haustorium (sometimes 2 haustoria) to appropriate the protoplasmic contents; haustoria pedicellate, the pedicel usually  $2-6\ \mu$  long,  $0.5-1\ \mu$  wide, abruptly enlarging and bifurcating successively 1 to 4 times and thus bearing 2 to 12 divergent assimilative branches  $2-12\ \mu$  long and  $1-2\ \mu$  wide. Sterigmata arising abruptly from procumbent hyphae at intervals frequently of  $3-25\ \mu$ , commonly  $1.5-4\ \mu$  high,  $1-1.5\ \mu$  wide at the base, tapering upward to a width of  $0.5-0.7\ \mu$  at the tip whereon is borne erectly a single conidium. Conidia colorless, usually composed of two parts: an upper empty membranous part generally  $4-13\ \mu$  long,  $0.8-1.2\ \mu$  wide proximally, gradually tapering upward, often more or less collapsed; and a lower filamentous living part tapering slightly toward both ends, commonly  $50-80\ \mu$  long and  $1.6-2\ \mu$  wide. Zygophoric hyphae often irregularly flexuous, sometimes winding about one another in some measure, both of them occasionally arising from separate mycelial filaments, but much more frequently only one arising from a mycelial filament, the other being supplied from a germinating conidium; zygosporangium at first smoothly subspherical and measuring  $8-11\ \mu$  in diameter, but its membrane at maturity collapsing loosely about the zygospore; the latter yellowish, subspherical,  $7-10\ \mu$  in diameter, boldly verrucose, having a wall  $1-2\ \mu$  thick that closely surrounds a spherical living cell  $4.3-6.5\ \mu$  in diameter.

Capturing and consuming two species of *Amoeba* commonly  $20-40\ \mu$  in width, it occurs in decaying leaves of deciduous trees (*Betula*, *Acer*, *Ulmus*) near Mercer, Wisconsin.

#### AN ACAULOPAGE DESTRUCTIVE TO A RETICULATE RHIZOPOD

Several soft maize meal-agar plate cultures which when well overgrown by *Pythium ultimum* had been further planted with small quantities of ash (*Fraxinus* sp.) leaves collected near Greeley, Colorado, early in October, 1945, showed after 35 days numerous ovoid bodies distributed sparsely over much of the agar surface, each curiously ornamented at its apex with a small tuft-like empty appendage. When the younger ovoid bodies, in which the appendage was still filled with protoplasm (FIG. 6, A, a), were examined closely they could often be seen attached basally to a hyphal branch that led backward to a scanty aseptate my-

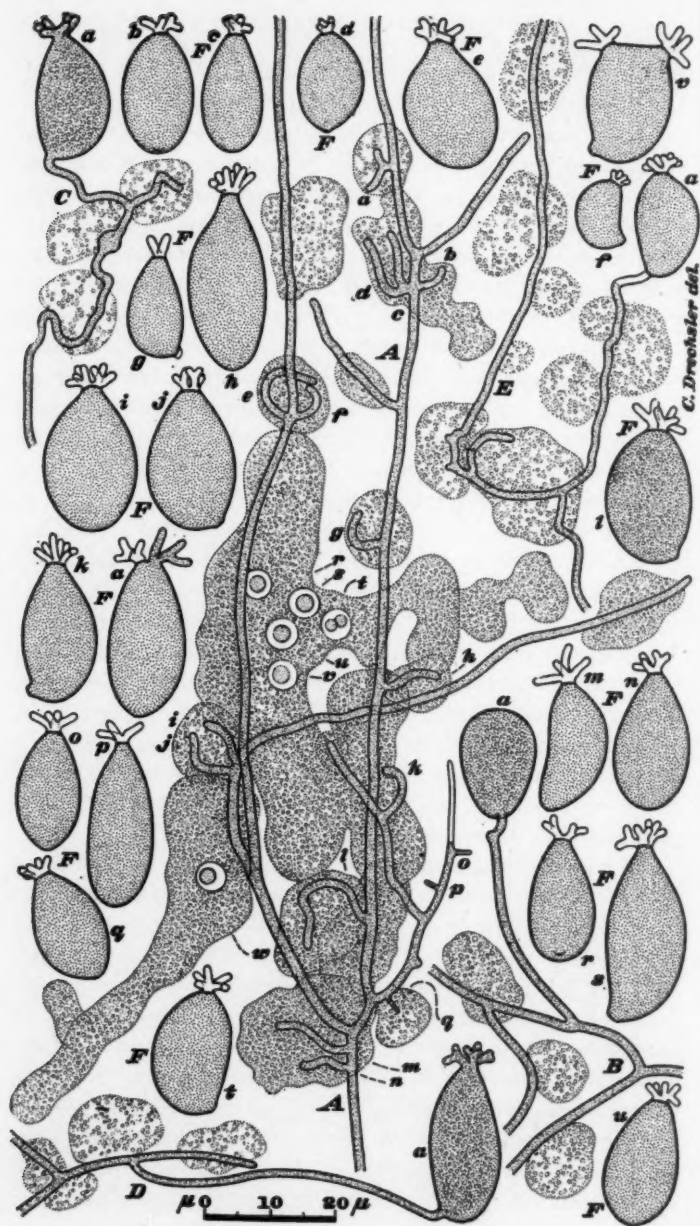


FIG. 6. *Acanlopaga crobylospora*.



celium composed of filaments mostly about  $1.3\ \mu$  wide. In following the longer hyphae through the soft agar substratum, branching was generally encountered at rather long intervals, but here and there, over stretches often 50 to  $100\ \mu$  or more in length, branches were given off in closer arrangement. As a rule the regions of such more copious ramification had an untidy appearance, owing to the presence of promiscuously scattered deposits of loose granular material (FIG. 6, *A*, *b*). On continued exploration an instance came to light wherein a ramifying mycelial tract (FIG. 7, *A*) was surrounded not with the usual messy deposits, but with an extensive mass of living protoplasm that kept on changing its shape continually through protrusion and retraction of pseudopodia. Nearby, in addition, several smaller protoplasmic masses, all having finely granular consistency like the main mass, from which they had evidently become separated, were found in irregular alignment along the hyphae. The smaller masses, together with the large mass, were invaded with short, frequently somewhat curved branches (FIG. 7, *A*, *a-n*) presumably functional as haustoria despite their meager outward differentiation. A few minute spurs (FIG. 7, *A*, *o*, *p*) about  $2.5\ \mu$  long and  $0.6\ \mu$  wide, that were found projecting from one of the longer branches, offered a little the appearance of adhesive organs, though it was not evident that they helped in holding fast the protozoan. As other modifications seemed lacking, it could only be presumed that escape of the animal was prevented by adhesiveness of the mycelial filaments themselves.

The captured protozoan was not surrounded by any pellicle—at least not by any pellicle thick enough to be clearly visible under the microscope. Its irregular shape and lack of integument gave ground for the suspicion that the untidy deposits of granular material could well have come from the disintegration of similar animals—a suspicion soon amply confirmed through the discovery of protoplasmic masses showing transitional stages of granular disorganization (FIG. 6, *B*, *C*; FIG. 7, *B-E*). Owing to the nearly normal condition of the actively struggling captive it was without difficulty recognized as being conspecific with numerous protozoans still at liberty in the culture. For the most part these protozoans were submerged in the soft agar, where they appeared as

FIG. 7. *Acaulopage crobylospora*.

three-dimensional networks measuring  $50\ \mu$  to 1 mm. across, and composed largely of anastomosing protoplasmic strands 2 to  $6\ \mu$  wide. At the periphery of the networks new strands were protruded rather briskly, and these, on anastomosing with one another, would form additional meshes; though simultaneously other meshes nearby might disappear through retraction of protoplasmic branches. As the multiple pseudopodial activity was not usually well coordinated the locomotion of the whole animal appeared rather slow, but nevertheless was fast enough for the species to spread throughout the culture. On the surface of the agar substratum the protoplasm seemed more inclined to collect in a thin sheet from which irregular arms would extend out, freely anastomosing here and there, to enclose lacunae of variable shape and extent. At the base of the pseudopodia the animal showed very little of the strongly acuminate modification illustrated in Leidy's (15: pl. 47, FIGS. 5-12; pl. 48), Penard's (16: 549, FIGS. 1-4), Cash's (2: pl. 8, FIGS. 3, 4), and Kudo's (14: 293, FIG. 134, e) figures of the reticulate *Biomyxa vagans* Leidy—a modification that Calkins (1: 350) cited as a distinguishing character of the genus *Biomyxa* in his key of the subclass *Proteomyxa* under the class Rhizopoda. The animal seems to fit better the description of *Leptomyxa reticulata* Goodey (13), a proteomyxan rhizopod originally described from soft agar plate cultures that had been inoculated with soil. It would appear to conform to the characterization of the genus *Leptomyxa* with respect to multinuclear condition; for in captured specimens plural nuclei (FIG. 7, A, r-w), measuring commonly 4.4 to  $5\ \mu$  in diameter and containing individually a slightly darker central body 2.3 to  $2.8\ \mu$  wide, seemed recognizable. In newly captured specimens, further, as also in free animals, multiple contractile vacuoles were distributed at varying intervals. The colorless, highly transparent character, and finely granular, almost homogeneous consistency of its protoplasm readily distinguished the animal from *Penardia mutabilis* Cash (2: 90-91), as well as from other colored genera compiled in the *Proteomyxa*.

In initiating asexual reproduction, hyphal branches little differentiated from other mycelial elements gave rise terminally to swellings that at first were of subspherical shape. After some

time, however, further enlargement took place mainly by elongation vertically into the air (FIG. 7, *B, a*), so that the swelling soon came to consist of an egg-shaped body with its long axis often nearly perpendicular to the supporting filament, its broadly rounded basal end resting in the agar, and its narrower end extended erectly into the air (FIG. 6, *A, a; B, a; FIG. 7, C, a; D, a*); the ovoid shape being conspicuously modified through apical prolongation of the body into a tuft of divergent processes having individually about the same width as the mycelial hyphae. Thereupon, as the protoplasm was withdrawn from the supporting hyphal branch and a basal retaining wall was laid down, the ovoid body became delimited as a conidium (FIG. 6, *C, a; FIG. 7, E, a*). Meanwhile the contents also of the distal processes were withdrawn backward, and a retaining wall was formed to delimit the living conidial cell from the empty membranous topknot. In some instances where the apical branches were relatively long— $5\mu$  being approximately their greatest length (FIG. 6, *D, a; FIG. 7, F, a*)—a cross-wall might be laid down in them to mark an intermediate stage in their evacuation. Now and then some of the branches would become evacuated while others still retained their contents (FIG. 7, *F, a*); and occasionally, again, the protoplasmic mass would be left extending slightly into the proximal portion of the topknot, so that plural retaining walls were needed to delimit two or more tubular elements separately (FIG. 6, *D, b, c*). Where a single retaining wall delimited the living cell distally in the usual manner (FIG. 6, *D, d-y; FIG. 7, E, b-u*) considerable diversity in the appearance of the membranous topknot, nevertheless, came about from differences in the number, length, and divergence of its constituent branches. In a few instances conidia of unusual width at the distal end were found ornamented with two well-separated tufts (FIG. 7, *F, v*).

Although the fungus is here described primarily from cultures prepared with Colorado material, its conidia have come under observation repeatedly for more than a decade in cultures planted with leaf mold and other kinds of decomposing vegetable detritus originating from different localities in Maryland, Virginia, Delaware, New York, Maine, and Wisconsin. However, owing to usually rather early evanescence of the branches bearing them,

their connection with a predaceous mycelium was not ascertained previously. A mistaken inference as to their probable identity was drawn from their fortuitous resemblance to frequently intermixed conidia produced by an apparently undescribed species of *Rhopalomyces* that often developed in the same soft agar plate cultures; the error being encouraged not merely by general similarity in size and shape, but also by the circumstance that the *Rhopalomyces* spores had their thin outer membrane extended emptily at either end. Yet the loosely protruding membrane of the *Rhopalomyces* spore was never seen disposed in a tuft; so that the predaceous fungus may conveniently be described under a specific epithet having reference to its curiously topknotted conidia.

***Acaulopage crobylospora* sp. nov.**

Mycelium sparsum; hyphis filiformibus, incoloratis, primum continuis, plerumque parce ramosis,  $1-1.8\ \mu$  (vulgo circa  $1.3\ \mu$ ) crassis, animalia minuta impediuntibus, deinde ramulos assumentes fere  $5-25\ \mu$  longos in eadem intrudentibus qui protoplasma magnam partem exhauriunt. Conidia in superficie materiae animalia ambientis sparsim oriunda, ex cellula viventi et crista vacua constantia: cellula viventi incolorata vel aliquantulum fumida, ovoidea,  $10.5-27\ \mu$  longa,  $6.8-14.3\ \mu$  crassa; crista vacua in  $2-15$  tubulis  $1-5\ \mu$  longis  $0.8-1.3\ \mu$  crassis consistens, saepius ad instar arbusculae divaricata.

Speciem *Leptomyxae* (forsitan *Leptomyxam reticulatam*) impediens necansque habitat in foliis *Fraxini* prope Greeley, Colorado, atque in humo silvarum et aliis materiis plantarum putrescentibus prope Beltsville, Maryland, et prope Georgetown, Delaware, et prope Webster, New York, et prope Presque Isle, Maine, et prope Butternut, Wisconsin, et in Arlington, Virginia.

Mycelium scanty; vegetative hyphae filamentous, colorless, at first continuous, for the most part only meagerly branched, measuring  $1-1.8\ \mu$  (commonly about  $1.3\ \mu$ ) in width, holding minute animals and intruding into them assimilative branches usually  $5-25\ \mu$  long which largely appropriate the protoplasmic contents. Conidia formed sparsely on the surface of the material surrounding the animals, colorless or slightly smoky, their single living cell usually of ovoid shape, mostly  $10.5-27\ \mu$  long and  $6.8-14.3\ \mu$  wide, bearing on its apex a frequently bush-like branching crest consisting of 2 to 15 empty membranous tubules  $1-5\ \mu$  long and  $0.8-1.3\ \mu$  wide.

Capturing and destroying *Leptomyxa* sp. (perhaps *Leptomyxa reticulata*) it occurs in decaying leaves of *Fraxinus* sp. near Greeley, Colorado, and also in deciduous leaf mold and other decaying plant

materials near Beltsville, Maryland, near Georgetown, Delaware, near Webster, New York, near Presque Isle, Maine, near Butter-nut, Wisconsin, and in Arlington, Virginia.

DESTRUCTION OF A RETICULOCYTE RHIZOPOD BY A  
PREDACEOUS HYPHOMYCETE

In a culture abundantly infested with the reticulate rhizopod habitually taken as prey by *Acaulopage crobylospora*, this animal was found being captured and destroyed by a septate mycelium be-longing presumably to a hyphomycete of the predaceous series most

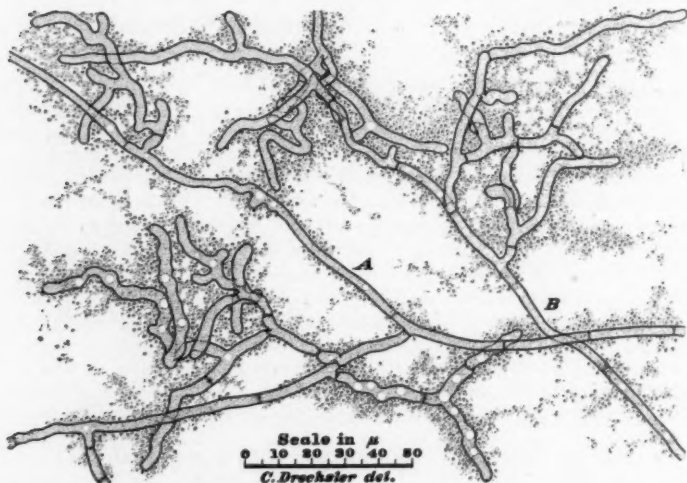


FIG. 8. Mycelium of a hyphomycete.

familiarly exemplified in *Arthrobotrys oligospora* Fres. On the longer filaments of this mycelium (FIG. 8, A, B) were borne here and there irregularly ramifying lateral branches. These branches were found surrounded by the same untidy granular deposits habitually associated with *A. crobylospora*, and they manifestly represented special assimilative elements despite their meager outward differentiation. Although the septate mycelium, with its constituent hyphae measuring about  $3\mu$  in width, was appreciably coarser than the continuous mycelium of the zoöpagaceous form, it yet bore



a striking resemblance to the latter—a resemblance obviously deriving from similarity in biological relationship to the same peculiar rhizopod. Unfortunately no reproductive bodies of any kind were produced whereby the specific identity of the septate mycelium might have been determined. A possibility worthy of consideration is that this mycelium might have belonged to one of the several described species, including notably *Dactylella atractoides* Drechsl. (11: 357–360), *Dactylella heptameres* Drechsl. (11: 352–354), *Dactylella rhombospora* Grove (6: 539–540), *Dactylella rhopalota* Drechsl. (11: 354–357), *Dactylella tenuis* Drechsl. (6: 538–539), and *Dactylaria pulchra* Linder (11: 349–352) which, though obviously referable taxonomically to the predaceous series of hyphomycetes, have not hitherto been observed in any important biological relationship to animals. At the time the species mentioned were tried out in cultures infested with nematodes and rhizopods of various kinds, the appearances usual in the destruction of non-pelliculate proteomyxan rhizopods were unknown to me, and therefore would very probably have been disregarded had they been present.

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## EXPLANATION OF FIGURES

FIG. 1. *Stylopage rhabdoides*; drawn to a uniform magnification with the aid of a camera lucida;  $\times 500$  throughout. A, Specimen of *Amoeba verrucosa* captured through adhesion to a meagerly branched mycelial filament from which a single haustorium has been intruded. B, C, Specimens of *A. verrucosa*, each captured through adhesion to a branched hypha from which two haustoria have been intruded. D, Specimen of *A. verrucosa* captured through adhesion to a branching hypha somewhat extensively enwrapping it; three haustoria have been intruded into the animal. E, Specimen of *A. verrucosa* held captive through rather extensive adhesion of mycelial hyphae; from these hyphae three haustoria have been intruded into the animal. F, Specimen of *A. verrucosa* captured through somewhat extensive adhesion to two branched hyphae, from each of which two haustoria have been intruded. G, Two specimens of *A. verrucosa*, a and b, huddled together and extensively enwrapped by hyphal elements belonging to three branching systems; seven haustoria have been intruded into a, six into b. H, Small specimen of *A. verrucosa* which after being captured through adhesion to a mycelial hypha, and subsequently being almost completely expropriated of its protoplasmic content by two rangy haustoria intruded into it, has succumbed to death.

FIG. 2. *Stylopage rhabdoides*; asexual reproductive apparatus drawn to a uniform magnification with the aid of a camera lucida;  $\times 1000$  throughout. A-D, Portions of prostrate hyphae from each of which has arisen an erect conidiophore, a, continuous at its narrowed distal end with a young conidium, b. E, Prostrate hypha from which has arisen an erect conidiophore, a, which is still continuous with the young conidium, b, being formed distally on it; from a has been extended a lateral branch, c, which is continuous with the young conidium, d. F, Portion of prostrate mycelial filament from which has arisen an erect hypha, a, that has given off a branch, b, which is still continuous with the young conidium, c, being formed terminally on it. G, H, Portions of prostrate hyphae, each with an erect conidiophore, a, that has become delimited from the conidium, b, borne at its tip. I, Portion of prostrate hypha from which have arisen three conidiophores, a-c, each delimited distally from the single conidium, d-f, borne terminally on it. J, Random assortment

of detached conidia, *a-z*, showing usual variations in size and shape. *K*, Conidium germinating.

FIG. 3. *Stylopage rhabdoides*; sexual reproductive apparatus drawn to uniform magnification with the aid of a camera lucida;  $\times 1000$  throughout. *A*, Two mycelial hyphae, *a* and *b*, that have put forth the sexual branches *c* and *d*, respectively, which have paired and are conjugating apically; *u*, place of union. *B*, *C*, Two young units of sexual apparatus; in each a conidium, *a*, and a mycelial hypha, *b*, have produced the sexual branches *c* and *d*, respectively, which through deposition of septa have formed gametangia that are conjugating apically. *D*, Two units of sexual apparatus derived from a conidium, *a*, and two separate mycelial hyphae *b* and *c*; the two sexual branches, *d* and *e*, arising as germ-tubes have been cut off to form gametangia which have conjugated severally with the gametangia formed on the sexual branches *f* and *g*, originating from the hyphae *b* and *c*. *E*, Two sexual units derived from a conidium, *a*, and a mycelial filament, *b*; the germ hyphae *c* and *d* have formed gametangia that have conjugated with others formed on the two sexual branches, *e* and *f*, extended from the filament *b*; a third sexual branch, *g*, given off by the filament *b* has attached itself as a supernumerary element to the conjugating pair *c* and *e*. *F*, *G*, Two units of sexual apparatus, each deriving from a conidium, *a*, and a mycelial filament, *b*; a gametangium supplied mainly, if not wholly, by the germ-tube *c* having conjugated with a gametangium borne on a sexual branch, *d*, from the mycelial hypha. *H*, Two sexual reproductive units derived from a germinating conidium, *a*, and two mycelial hyphae, *b* and *c*; gametangia supplied by two germ hyphae, *d* and *e*, have conjugated with gametangia borne on the sexual branches *g* and *h*, coming from *b* and *c*, respectively; the conidium has further put forth a third germ-tube, *f*. *I*, *J*, Two sexual reproductive units, each deriving from a conidium, *a*, and a mycelial hypha, *b*; in each unit a gametangium borne on a germ-hypha, *c*, has conjugated with one supplied by a sexual branch, *d*, coming from mycelial hypha *b*. *K-U*, Random assortment of mature zygospores showing usual variations in size, shape, and internal organization, as well as in disposition of membranous zygosporangial envelope. (*u*, Place of union between paired gametangia.)

FIG. 4. *Acaulopage ischnospora*; drawn at a uniform magnification with the aid of a camera lucida;  $\times 1000$  throughout. *A*, Portion of mycelium with two captured amoebae, *a* and *b*, into each of which a haustorium has been intruded; the captive *a* has a nucleus with about twelve flattened peripheral bodies. *B*, Portion of hypha with a captured amoeba having a nucleus of same structure as in *A*, *a*; a haustorium bearing four assimilative branches has been intruded. *C*, Portion of hypha with two captured amoebae, *a* and *b*, each containing a nucleus with a darkish central body; captive *a* shows an ingested fungus spore on the left side; captive *b* shows two similar ingested spores on the lower side and a smaller spore on the upper right side. *D-F*, Portions of hyphae, each holding captive a specimen of the same *Amoeba* sp. as that shown in *C*, *a* and *b*; a well developed haustorium is present in each animal. *G*, Detached conidia, *a-e*. *H*, Four conidia, *a-d*, attached to an *Amoeba* of the same species as *A*, *a*; the ypsiliform haustorium intruded by each conidium illustrates an early stage of development. *I*, Conidium that has captured a small amoeba, *a*, and has intruded a haustorium into it; a

germ-tube, *b*, is being extended from a distal position adjacent to the apical appendage. *J*, Two mycelial hyphae, *a* and *b*, that have supplied sexual branches with conjugating gametangia; a globose zygosporangium is being formed in the gametangium from *a*; *u*, place of union. *K-X*, Mature zygospores showing usual variations in size, shape, and internal organization, as well as in disposition of the surrounding membranous envelope.

FIG. 5. *Acaulopage ischnospora*; drawn at a uniform magnification with the aid of a camera lucida;  $\times 1000$  throughout. *A*, Portion of prostrate hypha with seven sterigmata, each bearing a (*a-g*) conidium that terminates in an empty appendage. *B*, Portion of prostrate hypha with a sterigma from which has been delimited a small, somewhat immature conidium still filled with protoplasm throughout. *C, D*, Portions of prostrate hyphae, each supporting erectly a mature, distally appendaged conidium. *E, F*, Portions of prostrate mycelial hyphae whereon are borne denuded sterigmata in numbers of nine (*a-i*) and seven (*a-g*), respectively. *G*, Somewhat immature detached conidia, *a-c*, filled with protoplasm throughout. *H*, Mature detached conidia, *a-p*, showing usual variations in size and shape, and in length of empty appendages. *I*, Mycelial hypha, *a*, that has produced two sexual branches, one of which has paired with a sexual branch from another mycelial hypha, *b*, while the other has paired with a sexual germ hypha from a detached conidium, *c*; *u*, place of union. *J-L*, Sexual reproductive units wherein a sexual branch from a mycelial hypha, *a*, has conjugated with a germ hypha coming from a detached conidium, *b*. *M*, Sexual reproductive unit likewise resulting from conjugation of a gametangium contributed by a mycelial hypha, *a*, with a gametangium supplied by a germinating conidium, *b*; *c, d*, cross-walls proximally delimiting the two gametangia; *e*, gametangium of mycelial origin in which the zygosporangium is developing; *u*, place of union. *N-S*, Mature zygospores showing usual variations in size, shape, and internal organization, as well as in disposition of the surrounding membrane.

FIG. 6. *Acaulopage crobrylospora*; drawn at a uniform magnification with the aid of a camera lucida;  $\times 1000$  throughout. *A*, Portion of branching mycelium surrounded with untidy deposits of granular protoplasmic detritus, *b*; connected to it is a full-grown conidium, *a*, whose apical processes are still filled with protoplasm. *B*, Portion of mycelium bordered with masses of degenerating protoplasm; connected with it is a full-grown but slightly immature conidium, *a*. *C*, Portion of mycelium bordered with masses of degenerating protoplasm; connected to it is a mature conidium, *a*, with empty apical appendage. *D*, Random assortment of detached conidia, *a-y*, showing usual variations in size and shape of the ovoid living cell as well as in make-up of the apical appendage.

FIG. 7. *Acaulopage crobrylospora*; drawn at a uniform magnification with the aid of a camera lucida;  $\times 1000$  throughout. *A*, Portion of branching mycelium with a captured specimen of *Leptomyxa* sp. (possibly *L. reticulata*); *a-n*, assimilative branches; *o-q*, narrow spurs perhaps having an adhesive function; *r-w*, nuclei of rhizopod. *B*, Portion of mycelium bordered with masses of degenerating protoplasm; connected to it is a young growing conidium, *a*. *C, D*, Small portions of mycelium, each bordered with several masses of degenerating protoplasm; connected to each portion is a full-grown conidium, *a*, with its apical outgrowths still filled with protoplasm.

*E*, Portion of mycelium bordered with masses of degenerating protoplasm; connected to it is a mature conidium, *a*, with empty appendage. *F*, Random assortment of detached conidia, *a-v*, showing variations in size and shape of the living ovoid cell, as well as in make-up of the apical appendage; in *a* three of the six branches in the appendage are still filled with protoplasm; in *v* two appendages are present on opposite sides of the flattened apical end.

FIG. 8. Mycelium of a hyphomycete distributed in an untidy expanse of granular detritus left after disintegration of a captured specimen of *Leptomyxa* sp. (possibly *L. reticulata*); two main mycelial filaments, *A* and *B*, are present, together with several irregularly ramifying assimilative branches attached to them.

## MORE NOTES ON GASTEROMYCETES<sup>1</sup>

S. M. ZELLER

(WITH 11 FIGURES)

This publication includes primarily some new species of *Calvatia* and *Scleroderma* and a review of certain American species of *Disciseta* and *Holocotylon*. Notes on a few other new or noteworthy species of Gasteromycetes comprise the remainder of the paper.

### *Arcangeliella lactarioides* sp. nov.

Fructificationes 2.5–3 cm. crassae, 1.5–2 cm. altae, agaricoideae, sphaeroideae demum expansae vel convexo-pileatae, disco leniter depressae, stipitatae; superficie glabra, innato-fibrillosa, sicca, pallide lutescenti, siccitate brunneola; peridio tenui, filamentoso, ductibus lactiferis praedito; columella percurrenti, lactiginosa, postremo stipitiformi, 4–6 mm. crassa; gleba alba, demum crenea, siccitate brunneola, inferne aperta, adnexa, ventricosa; locellis labyrinthiformibus, sporis albis partim impletis; septis albidis, ductibus lactiferis fartis; basidiis clavatis, quadrisporis, longis sterigmatibus munitis; sporis ellipsoideis, verrucosis (ut in *Russulis* *Lactariisque*), pedicellatis, 8–10.5  $\times$  6–6.3  $\mu$ .

Fructifications 2.5–3 cm. broad, 1.5–2 cm. high, resembling a stout agaric button, subspherical becoming expanded, convex pileate, somewhat depressed at summit, stipitate; surface smooth, innately fibrillose, dry, pale yellowish, drying brownish; peridium thin, especially below or at margins, where it breaks away from base of stem (columella), filamentous with lactiferous ducts; columella percurrent, becoming a stipe as the cap expands, 4–6 cm. broad, lactiferous; gleba white, becoming creamy, drying brownish, exposed below, adnexed, very ventricose, cavities labyrinthiform, partially filled with white spores; septa whitish in section, filled with lactiferous ducts; basidia clavate, four-spored, with long sterigmata bearing the spores acrogenously; spores ellipsoid, verrucose with protuberances of various sizes and somewhat connected by reticulate lines (as in *Russula* and *Lactarius*), pedicellate, 8–10.5  $\times$  6–6.3  $\mu$ .

<sup>1</sup> Published as Technical Paper No. 499, with the approval of the Director of the Oregon Agricultural Experiment Station. Contribution from the Department of Botany.

In fir woods below timberline (elev. 7500 ft.) in Diller Canyon, Mt. Shasta, California, July 27, 1940, *Wm. B. Cooke*, 14666, **type** (in Zeller Herb.).

This species is a clear connecting link between the *Gasteromyces* and *Lactarius*. It has spores like *Russula* and *Lactarius*, but the spores are borne acrogenously. Pine squirrels are fond of the fruiting bodies. The type was dried and no record of the color of the milk was made.

*CALVARULA EXCAVATA* Zeller (FIG. 2)

It was with some reluctance that the genus *Calvarula* was originally described<sup>2</sup> because the type collection is somewhat inadequate. Two additional collections at the Farlow Herbarium were taken in 1942 at Matheson Hammock, Dade county, Florida. Both of these contain numerous specimens in various stages of glebal development. The dried specimens, shrunk materially because of the gelatinous tramal peridium, are characterized by ridges over the surface indicating the location of the cortical plates. The surface becomes subgelatinous, drying avellaneous. The gleba is olivaceous, soft, with very thin fragile septa, which later break down into an olive-brown powdery mass, mostly spores. There is also a very slender, white columella which arises from the point of attachment and reaches somewhat beyond the center of the fructification. These two large 1942 collections verify our earlier opinion that *Calvarula* is a genus distinct from *Protophallus*.

The description of *C. excavata* may be emended to include "columella unbranched, white, very slight and thread-like, extending about to center of fructification"; and "basidia forming a palisaded hymenium with paraphyses and cystidia; cystidia long, conic, acute."

*CREMEOGASTER LEVISPORUS* Mattirollo

A critical study of the type of this genus and species (a portion of which is in Zeller Herb. comm. by C. G. Lloyd, July, 1924) has led the writer to agree with Fischer<sup>3</sup> that it is a good genus inde-

<sup>2</sup> *Mycologia* 31: 23-26. 1939.

<sup>3</sup> Fischer, Ed. Neue Beiträge zur Kenntnis der Verwandtschaftsverhältnisse der Gastromyceten. Ber. d. Schweiz. Bot. Ges. 45: 231-247. 1936 (see p. 232).

pendent from *Leucogaster* with which it was previously placed in synonymy.<sup>4</sup> It differs from *Leucogaster* in ellipsoidal, smooth spores which do not possess a gelatinous sheath.

The peridium has a black rind covering a hyaline layer of prosenchyma with isolated patches (islands) of large-celled parenchyma; the whole up to  $260\ \mu$  thick. The spores are  $6.2\text{--}8.75 \times 10\text{--}12.5\ \mu$ . Otherwise the structure is as Mattiolo<sup>5</sup> described it. The type specimen is very mature, not young as we previously stated.<sup>4</sup> This genus, along with *Leucogaster*, is one of the borderline genera between the *Melonogastraceae* and *Hymenogastraceae*. To include the two genera in *Melanogastraceae* as Mattiolo<sup>5</sup> has done is perhaps a little more satisfactory than to place them in *Hymenogastraceae*.

***Gelopellis hahashimensis* (S. Ito & S. Imai) Zeller n. comb.**

Syn. *Hysterangium hahashimense* S. Ito & S. Imai, Sapporo Nat. Hist. Soc. Trans. 15: 10-11. 1937.

Ito & Imai pointed out the close similarity of this species to *G. Thaxteri* from which it differs chiefly in the size of spores, color of gleba, and columella. The columella of *G. hahashimensis* as described and as illustrated is oddly pendent, piercing the gleba from the top of the fructification at a point opposite from the base and point of attachment. Such a structure must have evolved from a percurrent columella which in this later stage of development has entirely lost its attachment with the base of the fructification. Should this be considered an aberrant form or should one expect to find the highest form of gleba within the Hysterangiaceae-Gelopellaceae-Protophallaceae group to be that without conspicuous columella, perhaps in forms like *Calvarula*? The writer has previously assumed that the modification of the columella and amplification of sterile tissues in the fructification points to higher development, as exemplified by the origin of the Phallaceae and Clathraceae

<sup>4</sup> Zeller, S. M., and C. W. Dodge. *Leucogaster* and *Leucophleps* in North America. Mo. Bot. Gard. Ann. 11: 398. 1924.

<sup>5</sup> Mattiolo, O. Descrizione di una nuova specie italiana del genere *Cremeogaster* Mattiolo (*C. Klikae* nov. sp.) e considerazioni critiche sulla posizione sistematica di questo genere. Atti della R. Accademia della Scienze di Torino 69: 237-248. 1934.



within the Phallales, from families within the Hysterangiales (see *Mycologia* 31: 17-30. 1939).

*Holocotylon* Lloyd (emended)

Fructifications subglobose; sterile base none; peridium easily separable, fragile, dehiscing irregularly; gleba of small, empty chambers separated by very thin, delicate, fragile but persistent, tramal partitions; hymenium composed of a compact palisade or loosely grouped paraphyses, conidiophores, and basidia; capillitium none; spores spherical to ellipsoid, colored, mostly pedicellate, smooth.

This southern genus resembles *Arachnion* somewhat in outward appearance, but not in structure of the gleba, as Lloyd has pointed out.<sup>6</sup> Most of the species have conidia borne on branched conidiophores and the basidiospores are borne on sterigmata of various lengths. The basidiospores and the conidia of the individual species are quite similar in appearance; in fact, in some species, like *H. anomalum*, it is difficult to distinguish between them. Conidiospores have not been discovered in many Gasteromycetes. The writer has seen them in one other genus, the *Leucophlebs* stage of *Leucogaster*. In that genus, however, the conidial stage is not in the same fructification with the basidial stage, at least not together at the same time, as in *Holocotylon*. It is also of interest that in *Holocotylon* and *Leucogaster* the sterigmata are long and of various lengths on the same basidium.

The taxonomic position of *Holocotylon* is obscure. The dehiscence is like *Calvatia*, but since it is without capillitium and maintains structural characters of the gleba to maturity, it does not conform to the *Lycoperdaceae*. Although it is undoubtedly epigeous, the writer is inclined to place *Holocotylon* in the *Hymenogastraceae* because it retains glebal characters to maturity; there are usually some thicker tramal septa dividing the gleba, and *Holocotylon* is perhaps less stable than the genera of *Lycoperdaceae*. The morphological development and cytology of such little-known genera as *Holocotylon* must be studied before their taxonomic relationships can be better established.

<sup>6</sup> Lloyd, C. G. The genus *Holocotylon*. *Myc. Writ.* 2: 254-255. f. 94-96. Pl. 73, f. 5-8. April, 1906.

## HOLOCOTYLON BRANDEGEEANUM Lloyd (emended)

Fructifications depressed globose, up to 3 cm. broad; surface yellow, becoming bay brown, smooth, with adherent sand; peridium thin, fragile; gleba drying drab-gray (R) to drab (R), divided by an occasional major tramal septum; cavities empty; the usual tramal septa delicate, thin, fragile, white within, surfaces covered with chocolate to walnut brown (R) masses of spores; spores spherical, smooth, with one large oil globule, pedicellate, 5-6  $\mu$ .

In sandy soil.

SPECIMENS EXAMINED: MEXICO: Sinaloa, Culiacan, *T. S. Brandege*, **type** (in C. G. Lloyd Myc. Coll., No. 22748).

HOLOCOTYLON MEXICANUM Lloyd<sup>7</sup> (emended)

Fructifications subglobose to oblong, up to 15 mm.  $\times$  10 mm.  $\times$  8 mm.; surface white, smooth to somewhat cottony; peridium very thin, easily separable, leaving much of the gleba exposed at maturity; gleba buckthorn brown (R) to darker; cavities empty; tramal septa delicate, fragile, white within, surfaces covered with masses of light brown basidiospores; a few heavier whitish plates (similar in appearance to the peridium) dispersed through the gleba; hymenium loosely organized, composed of basidia and conidiophores; basidia in clustered groups over the surfaces of the tramal septa, four-spored, cylindrical, 8-10  $\times$  1.5-3  $\mu$ ; sterigmata slender, of various lengths, up to 18  $\mu$  long; basidiospores brown (almost hyaline under lens), smooth, ellipsoid, 3-5  $\times$  2.5-3.5  $\mu$ , sometimes pedicellate; conidia obovoid, 2.5-4  $\times$  2-3  $\mu$ .

In pastures or open places.

SPECIMENS EXAMINED: MEXICO, Tacuapan, *T. S. Brandege*, **type** (in C. G. Lloyd Myc. Coll., No. 22626); Michoacan, Morelia, *M. M. Solorzano* (in C. G. Lloyd Myc. Coll., No. 22746, under the herb. name, *Holocotylon rigidum*).

The specimens in the type collection are quite broken and fragmentary. A microscopic examination reveals appearances as of a puffball infected with some hyphomycetous fungus because the spores are borne on sterigmata of various lengths, and also because of the presence of branched conidiophores. The conidia are not pedicellate. The writer found three basidia floating free in a

<sup>7</sup> Lloyd, C. G. Myc. Writ. 2 (Letter No. 17): 1-2. Nov. 1, 1907.

microscopic mount from the type. The basidia were four-spored, with spores attached. The spores are borne on long sterigmata of various lengths, as in *Leucogaster*. In no other respect, however, is the fungus like *Leucogaster*. Some of the basidiospores to which the pedicels remain attached are similar to the spores of *Bovista*. The collection from M. M. Solorzano, Morelia, Mexico, is undoubtedly the one to which Lloyd referred on p. 271 and illustrated in fig. 113 (Myc. Writ. 2: 271. f. 113. July 1906). It is however the same as *H. mexicanum*, rather than *H. texense*, and the specimens are in fine condition.

*Holocotylon texense* Lloyd (emended)

Fructifications subglobose, somewhat depressed, 1.0–1.5 cm. broad, radicate by a rather stout rhizomorph; surface white, smooth, papery, somewhat areolated by the separation of the very thin, closely adnate, white exoperidium; peridium duplex, exoperidium as above, endoperidium papery thin, brown; gleba purplish brown at maturity; cavities empty; tramal septa papery thin, fragile, purplish brown at maturity, with two or three heavier tramal plates dividing the gleba from the base; basidia cylindrical forming a loose hymenium, four-spored; sterigmata slender, mostly near 12–18  $\mu$  long; basidiospores purplish brown, spherical, smooth (very slightly asperate as seen under oil immersion), long-pedicellate, 4–5  $\mu$ ; conidia subhyaline, subspherical to broadly ovoid, smooth, 3–4.5  $\mu$ .

SPECIMENS EXAMINED: TEXAS, Walker County, Huntsville, J. W. Stiles, **type** (in C. G. Lloyd Myc. Coll., No. 22747).

The type collection has become quite shattered (by insects?) and very little of the gleba is intact in most specimens. Basidia were found, however, with spores attached, and basidial characters indicate *H. mexicanum* and *H. texense* to be very closely related but yet quite distinct. The long-pedicellate basidiospores of *H. texense* are like those of a *Bovista*.

*Holocotylon anomalum* sp. nov.

Fructificationes circa 10 mm. altae, 7 mm. crassae, subglobosae vel turbinatae, rhizomorphis affixae; superficie alba, glabra, ochracea siccata; peridio tenuissimo, paene cartilagineo siccato; gleba olivacea; locellis parvis, irregularibus, vacuis; septis tramalibus brunneis, tenuibus, in angulis subscissilibus;

conidiophoro dichotomo-ramoso; conidiis ellipsoideis, subhyalinis,  $2.5-3 \times 3-4 \mu$ ; basidiis angusto-teretibus, hyalinis, longis sterigmatibus munitis; basidiosporis subglobois vel crasso-ellipsoideis, brunneis, levibus, longe pedicellatis,  $3.7-5 \times 3-3.7 \mu$ .

Fructifications about 10 mm. high, 7 mm. broad, subglobose to turbinate, with a distinct rhizomorphic attachment; surface white, drying tan, smooth; peridium very thin, drying almost cartilaginous; gleba olivaceous; cavities small, irregular, empty; tramal septa brown, thin, somewhat scissile where branched, lined on both sides by a distinct, crowded hymenium of large paraphyses and small basidia and conidiophores, basidia narrowly cylindrical, hyaline, with four long sterigmata; conidiophores mostly twice dichotomously branched; conidia ellipsoid, subhyaline,  $2.5-3 \times 3-4 \mu$ , basidiospores subglobose to short ellipsoidal, brown, smooth, mostly long pedicellate,  $3.7-5 \times 3-3.7 \mu$ .

On barren soil, Cisco, Eastland County, Texas, *E. A. Smith*, July 1938, **type** (in Zeller herb.).

Mr. E. A. Smith collected this species of *Holocotylon* along with a large collection (No. 1234) of *Disciseda Brandegeei*. The basidia are very narrow in this species and the conidiophores, which are twice bifid, appear very much like basidia with two long sterigmata which are once branched. Each conidiophore therefore bears four spores as do the basidia. Cytological studies to aid in determining the origin of basidiospores and conidiospores in *H. anomalum* might accordingly prove of value.

**HYSTERANGIUM STOLONIFERUM var. brevisporum var. nov.**

A typo differt: sporae  $10-14 \mu$  longae; fructificationes  $\pm 2.5$  cm. crassae.

As in the species, but larger, up to 2.5 cm. in diam., light tan becoming light brownish when dry, from a long (1-5 inches) radicate stolon; peridium  $380-420 \mu$  thick, composed of one layer of polygonal-celled parenchyma; gleba bluish when young, becoming brownish (drying buffy brown to light brownish olive), somewhat fragile; spores ellipsoid, light yellow,  $10-14 \times 5 \mu$ .

Under moss beneath vine maple, Trout Creek Forest Recreational Area, Linn County, Oregon, *S. M. Zeller*, May 21, 1938, **type**.

Whereas the spores of the typical variety are  $17-23 \mu$  long, these are but  $10-14 \mu$ . Otherwise it agrees with the typical variety in all respects but size.

**Leucogaster columellatus** sp. nov.

Fructificationes 1-2 cm. diam. siccatae, subglobosae vel reniformes; superficie levi, subviscida, pallide brunnea; peridio tenui, hyphis parallelibus, hyalinis, gelatinosis composito, intus duro luteoloque siccitate; columella hyphis magnis, parallelibus, hyalinis, gelatinosis composita, fragili rubroluteaque siccitate; gleba alba, siccitate pallide lutea cretaceaque; locellis parvis, subrepletis; basidiis subteretibus vel anguste clavatis, 2-4-sporigeris, sterigmatibus usque ad  $25\ \mu$  longis,  $1-1.5\ \mu$  diam.; sporis sphaeroideis, hyalinis,  $5-7.5\ \mu$  crassis, episporio lacunoso.

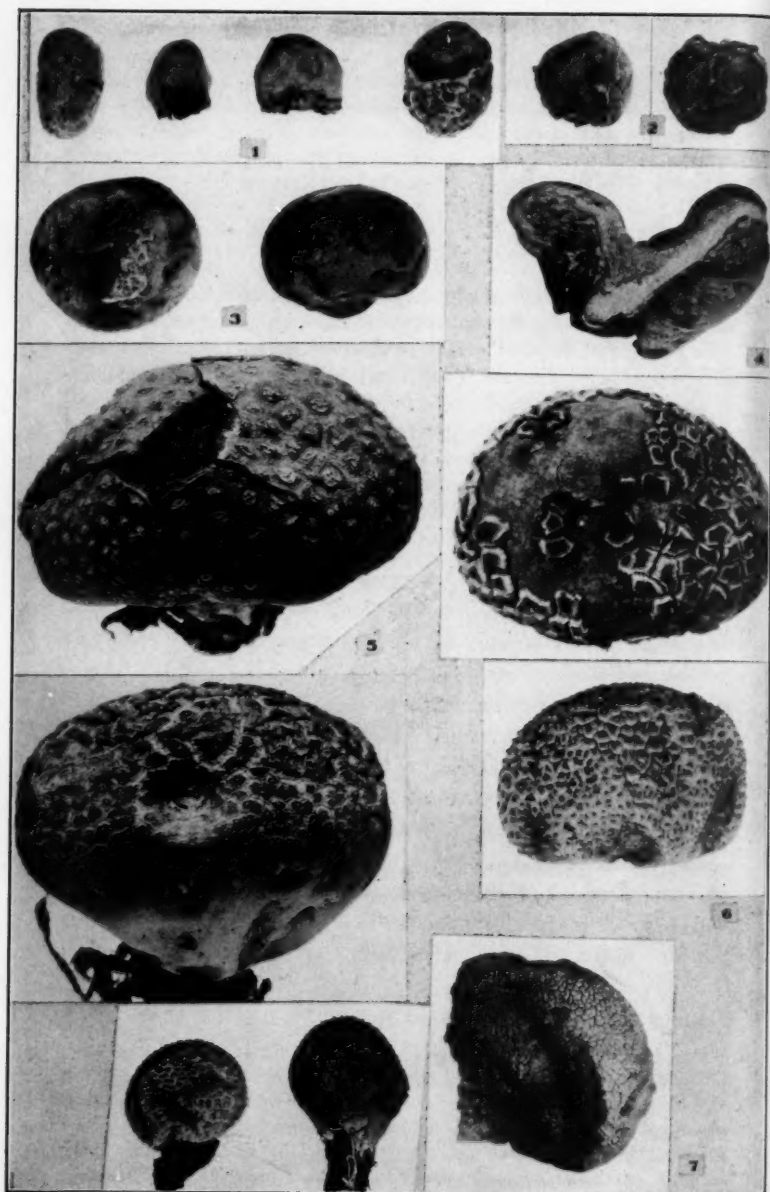
Fructifications 1-2 cm. in diameter (dry), subglobose to reniform; surface smooth, somewhat viscid with particles of humus adhering, light brown; peridium thin, of parallel, hyaline, gelatinized hyphae, drying flinty hard and quite amber in section; columella of large parallel, hyaline hyphae, gelatinized, drying brittle and reddish amber; gleba pure white, drying "light buff" (R), chalky texture; cavities small, almost filled with spores; basidia subcylindrical to narrow clavate, 2-4-spored; sterigmata of various lengths, up to  $25\ \mu$  long,  $1-1.5\ \mu$  in diameter; spores spherical, hyaline, surface pitted,  $5-7.5\ \mu$  in diameter.

Hypogeous under *Pinus ponderosa*, "Rim of the World," northeast of San Bernardino, California, May 1923, *N. L. Gardner*, No. 581, **type** (in Herb. Univ. of California, No. 233, 330, and small portion in Zeller Herb.).

This species is of special interest because of its columella. There are two fructifications in the type collection and in both the columella is very evident, reaching to the center of the fructification or farther. Because of the columella it may be that a new genus should be erected but all other characters are so typical of *Leucogaster* that it is placed in this genus, and the generic description may be amended to include species with a columella. This is perhaps as much justified as to include in *Lycoperdon* and other genera species with or without a sterile base.

**Leucogaster longisterigmatus** sp. nov.

Fructificationes subsphaeroideae vel oblongatae, 0.5-1 cm. diametro, usque ad 3 cm. long; superficie coactili hebetique, submarmorata, sordide brunneola vel atrobrunnea; fibrillis paucis, concoloribus; peridio  $130-200\ \mu$  crasso, hyphis periclinalibus compactis, prosenchymaticis constituto; gleba alba vel sordide griseola; locellis magnis, sporis repletis; basidiis quadrisporigeris, pyriformibus, sterigmatibus longioribus quam basidiis, usque ad  $25-35\ \mu$  longis;



FIGS. 1-7. Interesting Gasteromycetes.

sporis sphaeroideis vel subellipsoideis, crasse verrucosis, hyalinis, episporio utriculo gelatinoso hyalinoque, vestitis,  $7.5-10\ \mu$  ( $11.2-13.8\ \mu$  episporio incluso).

Fructifications subspherical to oblong, 0.5–1 cm. diameter, up to 3 cm. long; surface felty and unpolished, somewhat marbled in places where impressions of lacunae show through, dingy, brownish to blackish brown; fibrils not prominent, concolorous; peridium  $130-200\ \mu$  thick, of compact periclinal hyphae, approaching prosenchyma; gleba white to muddy grayish, lacunae large, filled with spores; basidia four-spored, with pedicels of various lengths, pyriform,  $12-19 \times 6-7\ \mu$ ; sterigmata longer than the basidia,  $25-37.5\ \mu$  long, stout; spores spherical or somewhat ellipsoid, coarsely verrucose, hyaline, enclosed in a hyaline, gelatinous sheath,  $7.5-10\ \mu$  ( $11.2-13.8\ \mu$  including sheath).

Under coniferous duff, Monument Peak, Linn County, Oregon, June 8, 1940, *Dr. Helen M. Gilkey* and *Dr. and Mrs. D. P. Rogers*, type.

The type collection fortunately includes specimens from 2 mm. diam. up. Even these smallest specimens have mature lacunae at the center, but just under the peridium, however, all of the lacunae are not wholly differentiated and indicate a morphological origin differing from that reported for *Leucogaster floccosus* Hesse by Fischer.<sup>8</sup> In *L. longisterigmatus* the long sterigmata are very distinctive, for in all other species so far described the spores are sessile or nearly so.

***Macowanites alpinus* sp. nov. (FIG. 4)**

Fructificationes usque ad 6 cm. crassae, agaricoideae; pileo in centro plano vel depresso, margine involuto; superficie levi, sicca, cremea vel ochracea; stipite brevi vel paene nullo, basi attenuato, paene albo vel cremeo, solido; gleba alba demum cremea, ventricosa, locellis magnis in centrum a peridiis vergentibus; peridio siccitate usque ad 4 mm. crasso, parenchymate spongioso composito, siccitate duro fragilique; basidiis quadrisporigeris, clavatis; sporis ellipsoideis, verrucosis,  $7.5-8.8 \times 6-6.5\ \mu$ .

Fructifications up to 6 cm. broad, agaricoid; pileus flat to depressed in center, margins incurved; surface smooth, dry, cream to buff shades when dry; stipe short-tapered below, almost wanting in some specimens; smooth, almost white, creamy, solid; gleba

<sup>8</sup> Fischer, Ed. *Mykologische Beiträge* 25. Jugendstadien des Fruchtkörpers von *Leucogaster*. *Mitt. Naturf. Ges. Bern.* 1921: 20–26. *illus.* 1922.



white, becoming cream color, ventricose, composed of anastomosing lamellae, forming large, labyrinthiform cavities; peridium (cap) very thick, up to 4 mm., dry, of spongy parenchyma, hard and brittle when dry; basidia four-spored, clavate; spores ellipsoid, verrucose,  $7.5-8.8 \times 6-6.5 \mu$ .

Under duff in fir woods, below Shasta Alpine Lodge in Cascade Gulch, up 7500 feet on Mt. Shasta, California, *Wm. B. Cooke*, No. 16797, **type**, Sept. 7, 1942.

***Secotium aurantium* sp. nov.**

Fructificationes solitariae, stipitatae, 3.5 cm. altae; pileo circa 1.4 cm. crasso, 1.5 cm. alto, apice rotundato umbonato, superne claro-aurantiaco, inferne capucino-luteo; peridio tenui, interne albo, inferne a stipite inseparabili; stipite tereti, usque ad 3 mm. crasso, glabro, externe albo, interne albo farctoque, inferne funiculis albis radicans, superne in columellam tenuem percurrentem procurrenti; gleba pallide brunnea, locellis parvis; hymenio paraphysibus crasso-clavatis basidiisque composito; basidiis crasso-clavatis, quadrisporigeris; sporis levibus, ellipsoideis, infra cicatricula sterigmatica ornatis, pallide brunneis,  $11-13 \times 6-7.5 \mu$ .

Fructification solitary, stipitate, whole plant 3.5 cm. tall, head almost 1.4 cm. broad, 1.5 cm. high, rounded umbonate above, bright orange above, capucine yellow at base; peridium not breaking from stem, thin, white within; stem terete, up to 3 mm. in diam., smooth, white without, white and stuffed within, from a white cord-like rhizomorph below, extended above into a narrow percurrent columella; gleba light brown, cavities small; hymenium of paraphyses and basidia; paraphyses broadly clavate; basidia broadly clavate, four-spored; spores smooth, ellipsoid, with a terminal sterigmal scar, light brown,  $11-13 \times 6-7.5 \mu$ .

On the ground, Mina Carlota, Sierra de San Juan, Trinidad Mountains, Cuba, *W. L. White*, No. 803, July 6, 1941, **type**.

The above collection consists of but one fructification which has a phalloid appearance because of the white cord-like rhizomorphs and general habit. The basidia and spores, however, are not like those of the phallaceous genera. Even though the spores and the general shape allow it to be included in *Secotium*, this species is placed there with considerable reservation, particularly until further collections may indicate more completely its morphological development.

*SECOTIUM MACROSPORUM* Lloyd, *Myc. Writ.* 1: 139. *pl.* 13, f. 12-16 (March) 1930.

EMENDED DESCRIPTION: Fructifications subglobose to obovoid, seldom pileate, sometimes turbinate, 2-3 cm. tall, 1-2 cm. broad, surface smooth to somewhat scurfy, white to grayish, then brownish at maturity; peridium papery, tough, thin; columella slightly projecting below, percurrent, tapering upward, up to 3 mm. in diameter below (dried), solid, white within; gleba mostly of partitions radiating outward and upward from the columella, wood brown, becoming almost sepia; cavities small; basidia pyriform, four-spored,  $12-15 \times 6-7.5 \mu$ ; spores brown, spherical, stout-pedicellate, one large oil globule,  $11-14 \mu$  in diameter, epispore thick, at first smooth, then reticulated by anastomosing shallow fissures.

TYPE LOCALITY: In open grass lands, May to June. Dallas, Texas.

This species appears in general habit, shape, size, color, and surface markings like *S. arizonicum* Shear & Griffiths but the spores are distinct in markings and are of a darker color, resulting in a much darker gleba. The spores have very stout pedicels. A collection from Dr. Roderick Sprague, Mandan, North Dakota, seems identical in characters with that from Texas (type locality).

#### ***Pompholyx occidentale* sp. nov.**

Fructificationes depresso-globosae, 3-7 cm. crassae, 2-5 cm. altae, superne albae demum sordidae vel purpureo-brunneolae, inferne levigatae vel rugosiusculae; peridio 4-6 mm. crasso, coriaceo, strato interno tenuissimo membranaceoque, comparate facile ab externo strato separabili siccato; gleba primo viridifusca vel vinaceo-brunnea, firma, demum obscure vinaceo-brunnea, pallide brunnea siccata, postremo pulvere sporum capillitiiq. brunnea repleta; septis albidis, postremo nonnumquam scissilibus; sporis sphaeroideis, obscure brunneis, echinulatis (echinis brevibus, truncatis),  $5-7.5 \mu$  crassis, basi breve pedicellatis vel hilis hyalinis munitis; capillitio obscure brunneo,  $5-7.5 \mu$  crasso. Odor foetidus.

Fructifications depressed globose, 3-7 cm. broad, 2-5 cm. high, white becoming sordid to purple brownish above, smooth, somewhat plicated into rounded ridges radiating from the point of attachment; peridium very thick, hard, leathery, 4-6 mm. thick, lined by a thin membrane which tends to adhere more to the gleba than to the peridium; gleba at first of a greenish or vinaceous brown color, firm, lacunae separated by whitish partitions, sometimes splitting along partitions at maturity, becoming dark vinaceous brown (R), drying lighter brown, a powdery mass of spores and capillitium; spores spherical, dark brown, echinulate with very short blunt spines,  $5-7.5 \mu$  in diameter, with a hyaline hilum or more

often short pedicellate; capillitium dark brown with heavy walls, 5–7.5  $\mu$  in diameter. Odor when fresh similar to flowers of Eastern skunk cabbage, *Symplocarpus foetidus*.

Hypogaeous becoming erumpent, under *Abies*, 5500–8000 ft. elevation on Mt. Shasta, August to September.

SPECIMENS EXAMINED: CALIFORNIA: Mt. Shasta, in west 40-acre lot at Horse Camp, *Wm. B. Cooke*, No. 14639, August 7, 1940; along Horse-Camp-Wagon-Camp Trail, *Wm. B. Cooke*, No. 15741, Aug. 18, 1941; and in Mud Creek Canyon above Mud Creek Dam, *Wm. B. Cooke*, No. 16671, July 2, 1942, **type** (in Zeller Herb.).

This species is placed in *Pompholyx* on the strength of Jaczewski's description<sup>9</sup> and rediscovery of *P. sapidum* Corda in Russia. In *P. occidentale* there is no evidence of nurse cells around the spores and the presence of a pedicel or a distinct hilum on the mature spore indicates this fungus should be included in *Pompholyx*.

Young fructifications were not seen but should be studied that more may be known of their morphological development and the origin of the basidia and the early glebal structure.

Specimens of *Pompholyx sapidum* Corda that Jaczewski studied, are now to be found in the Patouillard Herbarium at Farlow Herbarium, Harvard University. In this collection there are two or three stages of maturity. The largest piece is quite young. In it the spores are hyaline, smooth, 7.5–11  $\mu$  in diam., with a large hilum and sometimes even a portion of the sterigma still attached. In the darker fragment of glebal tissue the spores are dark brown, deeply reticulated with large meshes, 12.5  $\mu$  in diam. The older spores show no sign of a hilum or place of attachment. These spores, when stained, show no nurse cells, but the ragged appearance of some of them are quite like *Scleroderma* spores. The older piece of gleba could be *Scleroderma*.

Corda's illustrations of the spores of *Pompholyx sapidus* are difficult to explain without studying again the specimens he saw.<sup>10</sup> His figure 4 shows dark brown, verrucose spores with a peculiar reticulation dividing the spore into three equal parts. Fig. 5 shows the hilum surrounded by a larger white area, as though a part of

<sup>9</sup> Jaczewski, A. Bull. Soc. Myc. de France, 9: 169–173. 1893.

<sup>10</sup> Corda. *Pompholyx sapidus*. Sturm Deutschl. Fl. 19 & 20: 47–50. Tab. 15. 1841.

the episporium were torn away with the sterigma. If the episporium is torn away in this manner it seems incredible that the hilum would still show as a scar on the endospore, as Corda has it illustrated.

The specimens which Jaczewski studied do not have spores like those illustrated by Corda, but they are similar to those of *P. occidentale*.

The two known species of *Pompholyx* are aromatic and may or may not be hypogaeous. The peridium is harder or more leathery than in most species of *Scleroderma*, with which the genus has close affinities. In the mature spores of *Scleroderma* the hilum or any remains of the sterigma are wanting. Nurse hyphae or cells (Hülle) have not been found associated with the spores of *Pompholyx*. There is some doubt, however, that the spores of all species of *Scleroderma* are provided with nurse cells during the stages of maturation. Cunningham<sup>11</sup> has discussed this phenomenon to some extent, but there is some need for considerable study on the development of basidia and spores of echinulate-spored as well as reticulate- and verrucose-spored species of *Scleroderma*. What is the origin of the episporium markings? Do they all arise from the episporium or do some markings like reticulations originate from the nurse cells?

Cunningham<sup>11</sup> has mentioned *Pompholyx* as a synonym of *Scleroderma* (p. 277) but under his formal treatise on the genus he has omitted any reference to *Pompholyx*.

#### ***Scleroderma arenicola* sp. nov.**

Fructificationes subglobosae vel urceolatae, 2.5–6 cm. crassae; superficie glabra vel innato-fibrillosa, alba demum straminea vel luteola, siccitate pallide lutea vel sordide ochracea; peridio 1–3 mm. crasso, firmo, siccitate duro fragilique, apice stellatim vel irregulariter dehiscenti; gleba brunneola vel sepia demum atroinquinanti, siccitate fusca, postremo pulverulenta, sporis sphaeroidis, fusciculis, echinulatis reticulatisque, 15–22.5  $\mu$  diam.

Fructifications subglobose to urnshaped when stellately open at the top, 2.5–6 cm. in diameter; surface smooth or innate fibrillose, white becoming straw-color or yellowish, drying light buff to sordid ochraceous-buff, or with yellowish tints; peridium 1–3 mm. thick, firm, drying hard, brittle, dehiscing stellately at apex or rupturing

<sup>11</sup> Cunningham, G. H. The Gasteromycetes of Australasia XII. The genus *Scleroderma*. Linn. Soc. N. So. Wales, Proc. 56: 277–287. 1931.

irregularly; gleba brownish to sepia or blackish, drying fuscous, becoming powdery with remains of nurse-hyphae prominent; spores spherical, dark fuscous, echinulate with long sharp echinulae which tend to be ragged, not straight and stiff, large-meshed reticulations also visible through the dense echinulation, 15–22.5  $\mu$  in diameter.

In sand under *Pinus contorta* along the ocean coast, Waldport, Lincoln County, Oregon, August 4, 1928, *S. M. Zeller*, **type**.

This species is separated from the Friesian *Scleroderma Bovista* as described by Hollos chiefly in the larger size and the longer, sharper echinulae of the spores. This large-spored form among those with reticulate spores is essentially a northern and western species. This is undoubtedly the same species as that described by Coker and Couch as "*S. Bovista Fries*. Sense of Bresadola" and the size of spores of their Canadian citation (Canada. London. Dearness, No. 135B) compares well with the Oregon material.

***Scleroderma furfurellum* sp. nov.**

Fructificationes hypogaeae, subglobosae, 3 cm. altae, 2 cm. crassae, rugosissimae, siccitate durae; superficiei alba demum argillacea vel brunneola, superne squamis furfuraceis vestita; peridio duro, siccitate luteolo; gleba atra, e floccis albo-marmorata; sporis obscure fuscis, sphaeroideis, 10–12.5  $\mu$  diam., episporio crasso, leniter asperato.

Fructifications hypogaeous, subglobose, 3 cm. tall by 2 cm. wide, much wrinkled, drying hard; surface white becoming clay-colored or brownish, covered especially above with bran-like scales, scurfy; peridium hard, yellowish when dry; gleba black, marbled with white veins, some remains of nurse hyphae among the spores; spores very dark, spherical, thick episporium, slightly roughened, 10–12.5  $\mu$ .

In sandy soil, Perkinston, Stone County, Mississippi, *T. W. Brasfield*, No. 544, Oct. 20, 1939, **type** (in *Zeller Herb.*, communicated *W. H. Long*, No. 8465).

This species is characterized chiefly by the scurfy surface.

***Scleroderma subviscidum* sp. nov.**

Fructificationes epigaeae, subglobosae, 3–5 cm. crassae; superficiei glabra, subviscida, alba vel griseola, siccitate luteola; peridio 1–2 mm. crasso, lento duroque, prosenchymatico hyphis hyalinis, gelatinosis, compacte implicatis composito; gleba osseo-brunnea, siccitate concolori, compacta, pulverea, pos-

tremo e sporis et hyphis hyalinis vel coloratis, 4-7.5  $\mu$  diam., constanti; sporis 16-22  $\mu$ , sphaeroideis, obscure brunneis, uniguttulatis, tenuiter echinulatis; echinulis teretibus, truncatis.

Fructifications epigeous, subglobose, 3-5 cm. diameter; surface smooth, somewhat viscid holding particles of soil, white to grayish, drying with some yellowish tints; peridium 1-2 mm. thick, tough and hard, of somewhat gelatinized hyphae, tightly compacted into a hyaline prosenchyma; gleba bone brown, drying the same, compact, powdery, made up at maturity of spores and hyaline and colored hyphae 4-7.5  $\mu$  in diam.; spores spherical, 16-22  $\mu$  in diam., very dark brown, with a thick epispore, usually with one large vacuole, very finely echinulate; echinulae cylindrical with blunt rounded tips, 6 to about every 4  $\mu$ . 1.5  $\mu$  long, giving surface appearance of very fine beads.

On decayed granite soil, Prospect, Jackson County, Oregon, R. A. Pendleton, June 28, 1925, **type** (in Zeller Herb., 6842).

#### CALBOVISTA SUBSCULPTA Morse.

This genus is very close to *Mycenastrum* because of the type of capillitium but it is not like *M. corium* in peridial characters. Morse<sup>12</sup> did not report the species from Oregon but she reported it from Mt. Rainier only in Washington. *C. subsculpta* has been found in four locations in Oregon as follows: Baker County, Cornucopia, July 23, 1941; Clackamas County, on the trail to Paradise Park (Mt. Hood) from Twin Bridges Forest Camp, August 9, 1937; Klamath County, Crater Lake National Park, August 7, 1929; and Lane County, Fairview Mt., near Bohemia (*D. P. Rogers*), July 18, 1937. From Washington we have two collections by *Alex. H. Smith*, Nos. 14726 and 14854, from Mt. Angeles and Hurricane Ridge, both in the Olympic Mts., and one from Mt. Baker. Dr. Smith also collected it at Lake Fork Ranger Station, Idaho National Forest, Idaho, August 7, 1941.

#### CALVATIA CRETACEA (Berkeley) Lloyd (FIG. 7)

Lake Harbour, Baffin Island, Canada, J. Oughton, Aug. 10, 1939.

<sup>12</sup> Morse, E. E. A new puffball. *Mycologia* 27: 96-101. Pls. 12-15. 1935.

There were eleven fructifications of this interesting arctic species, each wrapped separately, but all bearing the above collecting data. The collections were communicated to the writer by Dr. H. S. Jackson, University of Toronto. This material from Baffin Island follows very closely the descriptions of the species furnished by Coker and Couch and by Lloyd, except that the capillitium measures  $3.7-7.5\ \mu$ . This then is in more perfect agreement with the collections from East Greenland which were described as *C. arctica* Ferdinandsen & Winge. No basidia or sterigmata were found in the Baffin Island specimens. This collection adds one more locality to the circumpolar distribution of *C. cretacea*.

***Calvatia subcretacea* sp. nov. (FIG. 6)**

Fructificationes depresso-globosae, 2-7 cm. crassae, 1.5-4 cm. altae, basi fibrillis albis gracillimis; exoperidio cretaceo albo vel fumosus, in areolas polygonias pyramidas circa 7 mm. crassas fissis demumque ex toto evanidis; endoperidio tenui furfuraceo, tenaci, melleo, primo integro dein irregulariter dehiscenti; basi sterili inconspicua; gleba primum olivaceo-lutea demum obscure umbrina, pulvere sporarum repleta; capillitio fracto, fusco,  $4-12\ \mu$  crasso; sporis sphaeroideis, levibus vel minute verrucosis, interdum breve pedicellatis, fuscis,  $3.7-6\ \mu$ .

Fructifications depressed globose, 2-7 cm. broad, 1.5-4 cm. high, base tapering to a point in young specimens, broad to almost flat when older, lightly attached by white mycelium; ectoperidium chalky white to smoke gray (R), conspicuous because of the polygonal pyramidal warts, the sides of which are mostly marked by lines which converge at the apex, sometimes with parallel lines at various heights around the warts, warts 7 mm. across or smaller, mostly pointed, seldom connivent, with darker apices than the sides, breaking away at maturity to expose a thin, firm, furfuraceous, dull chamois (R) to honey yellow (R) endoperidium, which breaks irregularly above; sterile base scanty or entirely absent, a soft olive buff (R) tissue of small cells; gleba passing through colors from olive buff (R) and drab (R) to a powdery burnt umber (R) mass; capillitium threads fragmented, often forked at about  $120^\circ$ , dark, undulating, sometimes, rough,  $4-12\ \mu$  in diam.; spores smooth or very finely verrucose, spherical, sometimes with short pedicels, slightly colored to rather dark,  $3.7-6\ \mu$ .

In duff of fir, hemlock, and spruce under alpine conditions from 6000 to 8500 feet elevation. Idaho, Washington, Oregon, and Northern California. July and August.



SPECIMENS EXAMINED: IDAHO: Boulder Lake (elev. 8300 ft.) near McCall, July 30, 1941, *A. H. Smith*, 15825; Lick Creek Summit, Idaho Nat. Forest, Aug. 2, 1941, *A. H. Smith*, 15930. (Both in University of Michigan Herbarium.)

WASHINGTON: Clallam County, Hurricane Ridge, Olympic National Park, July 7, 1939, *A. H. Smith*, 14860, Sol Duc Park, Olympic National Park, June 20, 1939, *A. H. Smith*, 14489. (U. of Mich. Herb.)

OREGON: Crater Lake National Forest (elev. 6000 ft.), *F. P. Sipe*, July 15, 1929, and July 22, 1943; Hood River County, above Cloud Cap Inn on Mt. Hood, *J. R. Kienholz*, July, 1936, *type*.

CALIFORNIA: Siskiyou County, Mt. Shasta, near Horse Camp (7500-8000 ft.), *Wm. B. Cooke*, Aug. 24, 1937, July 18, July 22, Aug. 12, 1938, July 8, 1940, July 29, Aug. 7, and Aug. 18, 1941, Nos. 8661, 10222, 10238, 10272, 14641, 15650, 15665, and 15742, respectively.

This alpine species has its closest affinities in the arctic species, *Calvatia cretacea*, but differs in several characters as follows: First, the color of the ectoperidium is essentially chalky white instead of creamy or buff, the warts are larger, smoky-dark at the summit, and somewhat contoured by lines (as in *C. arctica*), whereas in *C. cretacea* the warts are concolorous throughout. Second, the endoperidium is dull felty or furfuraceous instead of silvery smooth. Third, the sterile base is usually wanting in *C. subcretacea*.

#### CALVATIA CYATHIFORMIS (Bosc) Morgan

This species occurs across the country from the Atlantic to the Pacific. We have specimens from high up (7000 feet) in the Wallowa Mts. of eastern Oregon and from the Willamette Valley of western Oregon. Plants we have examined from such more eastern states as Maryland, Ohio, Missouri, and some collections from Texas conform quite closely to the usual original concept of the species. Throughout the west, however, there is considerable variability in the species from almost smooth-spored individuals through forms with very small finely asperate to coarsely verrucose spores. For instance, an extreme case is found in the University of Michigan Herbarium collection No. 1248 taken by *E. A. Smith*, Cisco, Texas, June 23, 1938. The specimens in this collection have the large stalk-like sterile base, the gleba is purplish brown, the spores, however, are finely asperate, short pedicellate, and measure  $2.5-4.3\ \mu$  including the sculpturing of the epispore. The capillitium is nearly hyaline and measures about  $3.75\ \mu$ . Externally the plants

agree with specimens of *Calvatia cyathiformis*, and we have referred them to that species, although with considerable reluctance. There seem, however, to be plenty of gradations of variability between this extreme variant and the typical forms.

*CALVATIA FRAGILIS* (Vitt.) Morgan

Coker and Couch have reported this species in northern states from Illinois to Wyoming and from Ontario, Canada. We have studied specimens of it from Texas and Oregon. The Texas collections are mostly the smooth form whereas those from Oregon have areolate peridia. It is the common purple-spored *Calvatia* of western Oregon. Several collections from Oregon were identified by C. G. Lloyd as *C. lilacina* var. *occidentalis* Lloyd, but he questioned whether it differs enough from *C. fragilis* to warrant the new name. The writer is convinced that the Oregon material is quite typically like that of European *C. fragilis*.

*Calvatia fumosa* sp. nov. (FIG. 10)

Fructificationes subglobosae vel depresso-globosae, 3-6 cm. crassae, leves vel leniter furfuraceae vel irregulariter rimosae, albae demum fumoso-griseae vel brunneae, basi funiculis albis radicante; peridio circa 1 mm. crasso, duplici sed partibus paene inseparabilibus ab utroque, intus cretaceo-albo, basi sterili vere absentibus vel parvulis; gleba pulverulenta, olivacea vel obscure brunnea; capillitio obscure brunneo, fracto, circa 6-18  $\mu$  crasso; sporis sphaeroideis, brunneis, verrucosis, breve pedicellatis, 5-7  $\mu$ .

Fructifications subglobose to depressed globose, 3-6 cm. in diameter, from a white cord-like rhizomorph; surface smooth to slightly furfuraceous under the lens, sometimes irregularly rimose, white then smoke gray (R) to hair brown (R); peridium drying about 1 mm. thick, of two layers almost inseparable but sometimes quite apparent, chalky white throughout except for the darker surface pellicle; sterile base wanting or merely thin and saucer-shaped, of small inconspicuous cavities with soft partitions; gleba becoming very powdery and completely falling away at maturity, passing through colors from Saccardo's olive (R) to mummy brown (R); capillitium dark brown, straight, unbranched, fragmented, sometimes rough, up to 6-18  $\mu$  thick; spores spherical, dark, strongly verrucose, short pedicellate, 5-7  $\mu$ .

Under conifers at altitudes of 5000 to 8000 feet on Mt. Shasta, California, and Crater Lake National Park, Oregon.

SPECIMENS EXAMINED: OREGON: Crater Lake National Park, *F. P. Sipe*, July 22, 1943, **type**.

CALIFORNIA: Siskiyou County, Mt. Shasta, near Everett Memorial Highway, June 27, 1941, *Wm. B. Cooke*, 15522; near head of Panther Creek meadows, July 28, 1941, *Wm. B. Cooke*, 15630; near Horse Camp, Aug., 1944, *Wm. B. Cooke*, 15778; and in Cascade Gulch below Shasta Alpine Lodge, Sept. 7, 1942, *Wm. B. Cooke*, 16799.

The color of this species is quite variable, some collections remaining almost entirely white. A single specimen from Spruce Cove, Trinidad, California, collected by *H. E. Parks*, Aug. 1933 is referred to this species with reservations. It has a much thinner peridium than the other collections. It may prove to be another species when a wide range of specimens are studied.

***Calvatia Lloydii* Zeller & Coker sp. nov. (FIG. 5)**

Fructificationes depresso-globosae vel subturbinatae, basi fibrillis albis radicante, 3-8 cm. crassae, superne tessellatae, cremeoalbae demum stramineae, vel rufobrunneae, inferne furfuraceae parum echinulatae; peridio duplici, sed stratis paene inseparabilibus, irregulariter dehiscenti; exoperidio tenui, areolato; endoperidio tenuissimo, papyraceo; basi sterili cellulosa, concava, purpureo-brunnea; gleba pallide olivacea demum obscure olivaceo-brunnea; capillitio brunneo, fracto, paulo ramoso, 2.5-6  $\mu$  crasso; sporis sphaeroideis, pallide brunneis, echinulatis, breve pedicellatis, 3-4  $\mu$ .

Fructifications 3-8 cm. in diameter, depressed globose to subturbinate, rounded or flattish above, pinched below and attached by ample, whitish rhizomorphs; surface at first creamy white, becoming stramineous or even reddish brown, dull or at times silky, areolated above into more or less prominent warts or obtuse spines, which are smaller and more or less furfuraceous below; outer peridium rather thin, and broken as described above, not readily separating from the inner peridium which is a papery layer so thin and delicate as to be very obscure and fading almost imperceptibly into the capillitium on the inner side; dehiscence irregular; gleba dusty, cottony mass of capillitium and spores changing from light to dark olive-buff; subgleba concave above extending somewhat in cup form up around the gleba, composed of small lacunae, the partitions thin and purplish brown, with a metallic luster; capillitium 2.5-6  $\mu$ , uneven, fragmented, somewhat branched, brown, with occasional linear pits; spores spherical, 3-4  $\mu$  in diameter, finely but distinctly echinulate, sometimes spines embedded in a clear envelope, often with torn remains of sterigma.

In dry coniferous duff of the Sierra Nevada Mountains of central California. May to August.

SPECIMENS EXAMINED: California; Stanislaus National Forest, July 28, 1931, *Frank Patty* (U. of Cal. Herb., No. 564470); Sequoia National Park, May 18, 1940, *Robert Wing* (U. of Cal. Herb. No. 698058); July, 1940, *Paul W. Colburn*, No. 4 (U. of Cal. Herb. No. 698060); Headquarters Area of Giant Forest, *Lee Bonar*, July 26, July 27, 1945 (U. of Cal. Herb. Nos. 698063 and 698064); along Halstead Creek trail, near Halstead Meadow, *Lee Bonar*, July 28, 1945, **type** (U. of Cal. Herb. No. 698059); Suwanee Grove, *Lee Bonar*, July 28, 1945 (U. of Cal. Herb. No. 698061); Tulare County, Giant Forest, *T. S. Brandegee*, Aug. 10, 1905, (Lloyd Herb. No. 7300 and U. of Cal. Herb. No. 564436); and Moro Rock Trail, Giant Forest, *Lee Bonar*, July 31, 1945 (U. of Cal. Herb. No. 698062). All of the collections are in the University of California Herbarium. There are parts of some of the collections in the University of North Carolina and the Zeller herbaria.

C. G. Lloyd saw the specimens collected by Brandegee in 1905 and he described them as a new *Calvatia*, but left the species unnamed. See Myc. Writ. 2: letter 18, p. 3. Jan. 1, 1908.) The specimens in the Lloyd Collections are under Catalogue No. 7300. Because Lloyd recognized the species as undescribed we are dedicating it to him, as *Calvatia Lloydii*.

In the fragmented capillitium with elongated pits and in the characters of the peridium, *Calvatia Lloydii* seems nearest *C. caelata*, as suggested by Lloyd. Aside from minor characters, it may be easily separated from that species by the absence of a massive plug-like base, the more slender capillitium with fewer pits, and the echinulate spores. In exterior appearance *C. Lloydii* and *C. tatrensis* Hollos are similar but they differ in size and surface of the spores as well as characters of the capillitium.

### ***Calvatia ochrogleba* sp. nov. (FIG. 11)**

Fructificationes turbinatae vel depresso-globosae vel irregulares, 7-9 cm. crassae, glabrae vel furfuraceae, sordide rufo-brunneae; peridio duplici sed stratis paene inseparabilibus, strato externo praetenui, strato interno crassiusculo, ochraceo; basi sterili fructificationis quartam inferam occupanti, centro convexula, alba vel subcaerulea; gleba subpulverulenta, firmula, primum lutea demum ochracea; capillitio hyalino, fracto, septato, 4-5  $\mu$ ; sporis albis, sphaeroideis, asperulis, 5-6.2  $\mu$ .

Fructifications turbinate to depressed globose, somewhat misshapen and lobed above, somewhat pointed below, 7-9 cm. broad and high; surface smooth to furfuraceous, dull reddish brown; peridium duplex, outer very thin, not easily separable, the two

layers breaking up together, inner medium thick, ochraceous; sterile base about  $\frac{1}{4}$  of fructification, convex above, cavities very small, whitish with bluish tints; gleba of very fine texture but not very powdery and not falling out easily, cavities very small, pale yellow-orange (R) to ochraceous-buff (R); capillitium hyaline, fragmented, septate, not pitted,  $4-5\ \mu$ ; spores creamy white, spherical, finely asperate,  $5-6.2\ \mu$ .

In meadow, Aurora, Marion County, Oregon, *Edwin Netter*, Sept. 18, 1941, **type**.

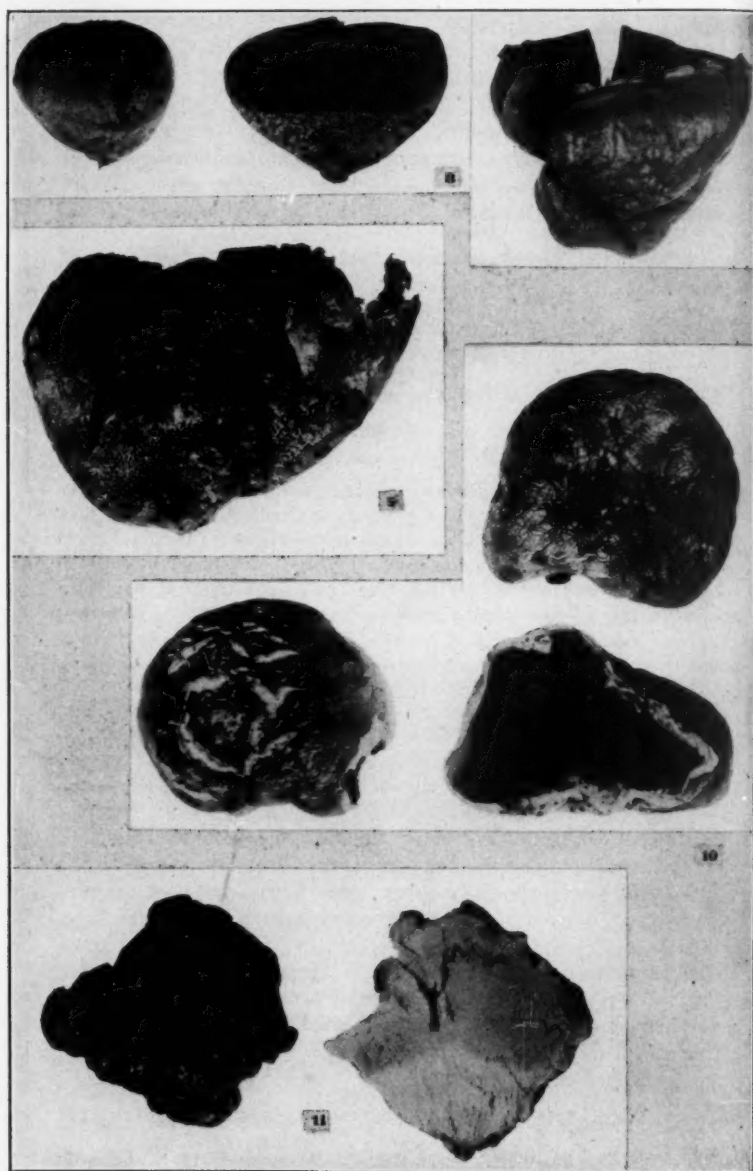
This is a very distinct species in many characters, especially in the color and texture of the gleba and the bluish color of the tissues of the sterile base.

***Calvatia rubrotincta* sp. nov.**

Fructificationes depresso-globosae, 4-7 cm. crassae, 3-4 cm. altae, inferne sulcatae, basi fibrillis albis radicante, inferne glabrae vel stipitatae vel furfuraceae, superne tessellatae, albae demum rubro-maculatae; peridio duplici, exoperidio albo, tenuissimo, evanido vel squamuloso persistenti, endoperidio crassiusculo fragili, irregulariter dehiscenti; basi sterili cellulosa, fructificationis decimam inferam occupanti, in centro paullo elevata, alba vel purpureo-brunnea; gleba pulverulentissima, benzo-brunnea vel obscure vinaceo-cinerea; capillitio obscure brunneo, paulo ramoso, fracto,  $3-4\ \mu$ ; sporis sphaeroideis, obscure brunneis, maxime verrucosis, breve pedicellatis,  $5-6.2\ \mu$ .

Fructifications depressed globose, 4-7 cm. broad, 3-4 cm. high, sulcate below, from a base with several white cord-like rhizomorphs; surface smooth to felty or furfuraceous below, areolated above by wrinkled reticulations, Hay's brown (R) when covered with spores, whitish where exoperidium has not disappeared, staining red in spots; peridium duplex, outer layer white, very thin, evanescent, or a mere flaky cover, inner layer somewhat thicker, brittle, easily broken up along the ridged reticulations; sterile base about  $\frac{1}{10}$  of fructification, convex above, soft, large-celled, white to purplish brown, metallic luster where torn; gleba very powdery, benzo brown to dark vinaceous-drab (R); capillitium dark brown, not pitted, not much branched, fragmented,  $3-4\ \mu$  in diam.; spores spherical,  $5-6.2\ \mu$ , dark, deeply verrucose, short pedicellate, many long pedicels floating free.

Under *Pinus ponderosa*, south of Silver Lake near Mt. Hager, Lake County, Oregon, *W. E. Lawrence*, July 23, 1934, **type**.



FIGS. 8-11. Interesting Gasteromycetes.

***Calvatia subpratense*** (Lloyd) Coker & Zeller n. comb. (FIG. 8)

Syn.: *Lycoperdon subpratense* Lloyd, Myc. Writ. 2: 231. pl. 62, f. 1-7. 1905.

Fructifications depressed globose or turbinate, bowl- or urn-shaped at maturity, 2.5-5 cm. high, 2-4 cm. broad; surface white, then pale ochraceous buff (R) or flesh pinkish, becoming shades of olivaceous or finally a dark shining metallic brownish where the inner peridium is exposed; outer peridium composed of small soft spines somewhat united at the apex, mixed with tiny simple spines and furfuraceous granules, which fall away from mature specimens; inner peridium thin, dark, inner surface at first densely felted with bases of capillitium threads but becoming shiny and metallic in old specimens, dehiscing by more or less stellate lobes which break up and fall away until the upper portion of the peridium has disappeared; sterile base prominent, of large cells, white, then brownish or purplish, often forming the whole lower half of the fructification, cup-shaped, separated from the gleba by a definite diaphragm, which is membranous and left smooth, shining, and metallic in old specimens; gleba white soft, becoming yellowish olivaceous, then light brownish olive; capillitium scanty at maturity, attached to the entire inner surface of the peridium and the diaphragm, pale brown, even or slightly irregular, up to 4  $\mu$  in diameter, sparingly branched, the walls moderately thick, not pitted, breaking up into short sections; spores spherical, smooth, dilute olivaceous, 3.2-4.7  $\mu$ .

In lawns and pastures. Massachusetts and Pacific Coast states.

SPECIMENS EXAMINED (by one or both of us): MASSACHUSETTS: Essex County, Peabody, *R. B. Mackintosh* (C. G. Lloyd Cat. No. 13207); Middlesex County, Cambridge, *A. B. Seymour*, **type** (C. G. Lloyd Cat. No. 51437). WASHINGTON: Klickitat County, Bingen, *W. N. Suksdorf* (in C. G. Lloyd Coll.). OREGON: Benton County, Corvallis, *H. C. Gilbert*, Nov. 19, 1915 (in O.S.C. Herb. 7146), *Roderick Sprague*, Oct., 1930 (in O.S.C. Herb. 7792), *S. M. Zeller*, Aug. 9, 1919, Oct. 19-25, 1937, July 7, Aug. 14, Nov. 17, 1939, May 8, 1940; Clackamas County, Canby, *S. M. Zeller*, Oct., 1926; Douglas County, Glide, *S. M. Zeller*, June 14, 1937; Marion County, *S. M. Zeller*, Oct. 14, 1926; Multnomah County, Gresham, *S. M. Zeller*, Nov. 18, 1927; Yamhill County, Newberg, *S. M. Zeller*, Oct., 1926; Washington County, Scholls, *S. M. Zeller*, Sept., 1937.

*Calvatia subpratense* is the most common puffball occurring in lawns and pastures in western Oregon. It has a tendency to grow in "fairy rings" in lawns, stimulating the growth of grass in the



circumference of the rings but not doing the damage to grass occasioned by *Marasmius oreades*.

This species, as already stated by C. G. Lloyd, answers quite well the description of *Lycoperdon pratense* Pers. but until such time as we may be able to prove just what Persoon's *L. pratense* is, it seems best to us to retain Lloyd's specific name for the American plant but to transfer it to the genus *Calvatia*.

It is puzzling what becomes of the capillitium during maturation, as it is definitely scanty in the ripe condition although the peridium and diaphragm show a dense coating of capillitium bases. It must be that the bases of the threads are less subject to autolysis than the main portion of the threads after they break away.

**Abstoma Townei** (Lloyd) n. comb.

Syn. *Catastoma Townei* Lloyd, Myc. Notes 67: 1168. July, 1922; 68: 1175. pl. 228, fig. 2337. Jan., 1923.

EMENDED DESCRIPTION: Fructifications subglobose, drying 2.5 cm. broad and 3 cm. high, universal veil dull drab, flocculent, breaking up as in *Amanitas*, leaving small flecks and patches over the surface of the ectoperidium which otherwise is smooth, very slightly viscid, Sanford's brown (R) to slightly lighter, thin, brittle, breaking away as in *Bovista*; endoperidium thin, whitish; gleba with tints of yellow and purplish when young, firm and compact though pulverulent; capillitium thin-walled, tinted slightly yellowish; spores spherical, deeply verrucose, purplish brown, pedicellate, 7.5–12.5  $\mu$  in diameter.

On ground. California.

SPECIMENS EXAMINED: **Type** collected by Stuart S. Towne (in Lloyd Myc. Collections, No. 31027).

The type collection consists of one young specimen, which is entirely inadequate for diagnosis, but since it bears a name, an effort should be made to retain it if possible until adequate material of the species is discovered. As Lloyd suspected, it does not belong in *Catastoma*. It undoubtedly belongs in *Abstoma* although the universal veil is not as much of a "sand case" as found in some other species of this genus.

The upper part of the gleba matures first and the only mature portion in the type specimen is centrally located in the top part. It

is purplish and is surrounded by a yellowish, less mature portion. The basal portion is still whitish and made up almost entirely of capillitium, much as that observed in *Disciseda Uplandii* (Lloyd) Zeller.

***Disciseda ater* (Lloyd) n. comb.**

Syn. *Catastoma ater* Lloyd, Myc. Writ. 5: Note No. 53: 756-757. f. 1131. Feb. 1918.

EMENDED DESCRIPTION: Fructifications subglobose, 3 cm. in diameter (dry), black; ectoperidium black, shiny, very thin, breaking away irregularly leaving the light-colored endoperidium exposed in spots or angular patches; endoperidium dull, light clay color, thickish and of the consistency of an oak gall, dehiscing by a large, slightly mammosse, apical stoma; gleba pulverulent, mummy brown (R); capillitium brownish,  $3.7\text{--}5\ \mu$  in diameter, irregular, heavy walled, fragmented, slightly branched; spores subglobose to ovoid, apiculate, brown, smooth to asperulate, one large vacuole,  $3.75\text{--}5\ \mu$  in diameter.

In a dry gravel wash, San Gabriel watershed, Cow Canyon, Los Angeles County, California, Sept. 17, 1917, *I. M. Johnston*, No. 2, **type** (in Lloyd Myc. Collection, No. 33650).

It is unfortunate that throughout Lloyd's description and notes on this species he has referred to both the ectoperidium and endoperidium as "endoperidium." It is only by examination of the type specimen that one may straighten out his notes. It is hoped this is adequately corrected in the above emended description. Surely Cunningham did not have access to the type collection when he erroneously placed *D. ater* in synonymy with *D. candida*. There is nothing about the two that is similar and they could hardly be confused. The fructification of *D. ater* has the general appearance of a black weathered oak gall. There is no sand case; in fact, the ectoperidium is a closely adhering, thin, shell-like layer.

***Disciseda Brandegeei* (Lloyd) n. comb. (FIG. 3)**

Syn. *Catastoma Brandegeei* Lloyd, Myc. Writ. 5 (Letter 65): 7. March, 1917; 6: 897-898. pl. 137. f. 1576. Oct. 1919; 7: 1168. July 1922.

EMENDED DESCRIPTION: Fructifications globose or depressed globose, 1.5–3.5 cm. in diam.; exoperidium thin, peeling away from the endoperidium in thin shell-like flakes, not forming a cup, smooth and with a silky sheen, or drying dull, usually pale or sordid whitish, but sometimes with buff or even brownish tints; endoperidium thin, smooth, deep olive-buff (R) or with pale slaty appearance, sometimes with lacerated plane aperture below at point of attachment of fructification; gleba at first white to smoky white then wood brown (R), avellaneous (R), buffy brown (R), or even Dresden brown (R) at maturity; capillitium pale to yellowish brown, slightly branched at right angles, broken, 2.5–5  $\mu$  not pitted; spores globose, 5–8.6  $\mu$ , brown, smooth.

In open areas in dry barren soil. Summer. Texas to California and Mexico.

SPECIMENS EXAMINED: MEXICO: Sinaloa, Culiacan, T. S. Brandegee, 1904, **type** (in C. G. Lloyd Myc. Coll. No. 37840); Texas, Eastland County, Cisco, E. A. Smith, July and August, 1938, Nos. 1210, 1218, 1234, 1237 (in Univ. Mich. Herb.).

The type collection is much darker brown than most of the Texas material, which numbers about 90 specimens. The dark exoperidium of the type collection, containing one specimen, is of such a dark brownish color as to suggest it might have been too severely handled or not naturally dried.

The basal dehiscence of the endoperidium is seldom apparent. The basal rhizomorph originates in the endoperidium, thus penetrating the ectoperidium. When the rhizomorph breaks away from the endoperidium it may or may not leave an aperture to the gleba. There is no indication of an apical stoma.

The one other species with smooth spores, *D. levispora*, is quite different from *D. Brandegeei*, in size of spores and peridial characters.

**Disciseda defodiodis** (Lloyd) n. comb. (FIG. 1)

Syn. *Calvatia defodiodis* Lloyd, Myc. Writ: 4 (Letter 44): 8. Jan., 1913.

Fructifications oblong to turbinate, usually truncate above, 2–3 cm. high, 0.8–2 cm. broad, with a rooting rhizomorph; sterile base none; peridium duplex; outer layer thin, white, soft, of loosely interwoven hyphae, usually left as a cup around the lower half of

the fructification; inner layer thin, but hard and white like the shell of reptilian eggs, sometimes hard when dry, tough but splitting most readily along vertical lines, dehiscing circumscissilely or by ruptures radiating from the basal point of attachment after liberation from the ectoperidial cup; gleba pulverulent, pale olive, drying deep olive buff (R); capillitium cobwebby, branched, flaccid, subhyaline, thin-walled, rarely with rounded protuberances, much broken,  $3.75-5\ \mu$  in diam.; spores subglobose to ovoid, sometimes somewhat angular, subhyaline, smooth, rarely very finely asperate under high power lens,  $3.75-5\ \mu$  in diam.

Imbedded in soil with top about even with surface of ground. Late June to summer, North Dakota and Wyoming.

SPECIMENS EXAMINED: NORTH DAKOTA: Lamoure County, Kulm, J. F. Brenckle, June 24, 1923 (in C. G. Lloyd Myc. Coll., No. 22362); Morton County, near Mandan, R. Sprague, June 10, 1942 (in Zeller Herb.). WYOMING: Park County, Meeteetse near Cody, Simon Davis, **type** in C. G. Lloyd Myc. Coll., No. 22361).

This species is an oddity in that an unbroken endoperidium might at first be mistaken for the egg of a reptile. It recalls to the writer's mind the occasion when he received from a botanical colleague reptilian eggs for identification as a gasteromycete.

***Disciseda Johnstonii* (Lloyd) n. comb.**

Syn. *Catastoma Johnstonii* Lloyd, Myc. Notes **61**: 898. Oct., 1919; **67**: 1168. July, 1922.

EMENDED DESCRIPTION: Fructifications depressed globose, about 1.5 cm. in diam.; ectoperidium thick, flaking away leaving a mere disk at the base, dark brown, rough; endoperidium thin, pliable checked by tiny cracks into angular areas, dehiscence not evident; gleba reddish brown, powdery, flocculent; capillitium very light yellow, wavy, with heavy walls, somewhat branched, broken,  $3.5-4\ \mu$  in diameter; spores globose to angular, verrucose,  $4-5\ \mu$ , yellowish, sometimes short-pedicellate.

At side of trail in chaparral belt, upper Sonoran Zone, alt. 3000 feet, Cucamonga Canyon, I. M. Johnston, No. 238, Nov. 10, 1918, **type** (in Lloyd Myc. Collection, No. 33647).

Lloyd described the spores as smooth although in the type they are quite verrucose, at least much more so than are those of *D. candida*.

***Disciseda levispora* (Lloyd) n. comb.**

Syn. *Catastoma levisporum* Lloyd, Myc. Notes 59: 853. f. 1428. 67: 1168. July, 1922.

EMENDED DESCRIPTION: Fructifications 2.5–3 cm. in diameter, subglobose; sterile base none; peridium firm cartilaginous; ectoperidium thick, firm, smooth, dark brownish to blackish, not easily separable from the endoperidium which is papery thin, whitish, dehiscing almost stellately at the apex; gleba dark purplish, brown, powdery and flocculent; capillitium wavy, heavy walled, pale brown, slightly branched at right angles, broken, 6–8  $\mu$  diam.; spores spherical, smooth, 3.5–4.5  $\mu$ , short pedicellate.

On ground in vacant lot under pepper trees, Claremont, California.

SPECIMENS EXAMINED: CALIFORNIA: Los Angeles County, Claremont, Lois M. Clancy, Oct. 9, 1916, **type** (in Lloyd Myc. Collection, No. 8782).

The type collection is quite inadequate, being the fragmentary portion of one fructification. It is hoped the collection reported later by Lloyd as taken by I. M. Johnston (Myc. Notes 67: 1168. 1922) is more representative.

***Disciseda luteola* (Lloyd) n. comb.**

Syn. *Catastoma luteolum* Lloyd, Myc. Writ. 2: Letter 11: 4, June, 1906; 7: 1168. July, 1922.

EMENDED DESCRIPTION: Fructifications depressed globose, about 1.5–2 cm. broad, drying stramineous to clay-color; ectoperidium very thin, furfuraceous, fugacious; endoperidium thin, flaccid, papyraceous, buffy to cream color below, light brownish above, dehiscing at basal point of attachment through torn aperture; gleba buffy brown (R) to olivaceous, pulverulent; capillitium irregular, heavy walled, very slightly branched, pale yellow, fragmented, about 3–4  $\mu$ ; spores spherical, almost smooth but somewhat verruculose, apiculate, 5–6.5  $\mu$ .

On the ground. Tahoe Lake, California. July.

SPECIMENS EXAMINED: CALIFORNIA: Lake Tahoe, P. B. Kennedy, July, 1905, **type** (in Lloyd Myc. Collection, No. 37839).

Lloyd's observation that this species is close to *Disciseda hypogaea* (Cooke & Massee) Cunn. is accepted but the two are quite

distinct in the episore markings and in the dehiscence of the endoperidium.

***Disciseda uplandii* (Lloyd) Zeller n. comb.**

Syn. *Catastoma Uplandii* Lloyd, Myc. Notes **61**: 897. pl. 137 f. 1575, Oct., 1919; **67**: 1168. July, 1922.

EMENDED DESCRIPTION: fructifications depressed globose 1.5–2 cm. in diameter, purplish brown when dry; exoperidium thin, rough with adherent sand, persisting over the lower half of the fructification; endoperidium thin, papery, slaty-brown, dehiscing by a torn apical pore; gleba buffy brown (R) to drab (R), pulverulent, nearly all capillitium in lower part, with very few spores, spores plentiful above; capillitium irregular, wavy, somewhat branched at right angles without septum, walls heavy, slightly pitted, brown, 3–4  $\mu$  diameter; spores spherical, 6–7.5  $\mu$ , dark brown, verrucose, with short remains of the sterigma which is proportionately broad (at least 1.2–1.7  $\mu$  broad).

In open grassy ground, altitude 1200 feet, Upland, California. December.

SPECIMENS EXAMINED: CALIFORNIA: San Bernardino County, Upland, I. M. Johnston, Dec. 25, 1918, No. 286 **type** (in Lloyd Myc. Coll., No. 8785).

This species is characterized by the very rough, verrucose spores, which have a short but conspicuously broad pedicel or apiculus and by the tendency toward a sterile base. Lloyd's "very minute apiculus" was not found. The capillitium is very compact in the lower part of the gleba with very few if any spores, except those that accidentally moved in as the fructification was vertically sectioned. Some microscopic mounts from this part of the fruiting body had no spores in them. This sterile basal portion made up entirely of capillitial filaments is novel among the Lycoperdaceae.

*D. uplandii* also differs from other species of the genus in the pitted capillitium, which Cunningham<sup>13</sup> states definitely is not a character of *Disciseda*. This anomalous species recalls somewhat the gleba of the Mesophelliaceae<sup>14</sup> and the Geastraceae.

<sup>13</sup> Cunningham, C. H. Gasteromycetes of Australia and New Zealand. John McIndoe, printer, Dunedin, N. Z., 1942 (see pp. 135–136).

<sup>14</sup> Zeller, S. M. Representatives of the Mesophelliaceae in North America. Mycologia **36**: 627–637. *illus.* 1944.

## EXPLANATION OF FIGURES

(All specimens except fig. 11 natural size)

FIG. 1. Four fructifications of *Disciseda defodioidis* (Lloyd) Zeller. One and four show the ectoperidium broken away exposing the endoperidium above. Two and three are endoperidia broken from the basal portion by circumscissile rupture.

FIG. 2. Two dried fruiting bodies of *Calvarula excavata* Zeller in which the location of sutures of the tramal peridium is revealed as ridges.

FIG. 3. Two specimens of *Disciseda Brandegeei* (Lloyd) Zeller illustrating the rupture of the exoperidium. In the first specimen the ectoperidium is torn away exposing the basal pore in the endoperidium.

FIG. 4. Median section of a dried specimen of *Macowanites alpinus* Zeller.

FIG. 5. *Calvatia Lloydii* Zeller & Coker. Specimen above shows irregular dehiscence; specimen below somewhat less mature.

FIG. 6. Two fructifications of *Calvatia subcretacea* Zeller. Specimen above shows dehiscence of both the ecto- and endoperidium. Specimen below illustrates younger stage and character of the ectoperidium.

FIG. 7. Three specimens of *Calvatia cretacea* (Berkeley) Lloyd illustrating peridial characters in comparison to those of *C. subcretacea* (fig. 6).

FIG. 8. Three fructifications of *Calvatia subpratense* (Lloyd) Coker & Zeller, the first showing irregular apical dehiscence of a small specimen; the second, a median vertical section showing gleba and sterile base; the third, an old overwintered specimen.

FIG. 9. *Calvatia tatrensis* Hollos (see Mycologia 33: 213. 1941).

FIG. 10. Three specimens of *Calvatia fumosa* Zeller showing the smooth peridium, its irregular cracking, and gleba with little or no sterile base.

FIG. 11. Exterior and interior of *Calvatia ochrolebda* Zeller.  $\times \frac{1}{2}$ .



## FUNGI CAUSING DECAY IN WOODEN BOATS

ROSS W. DAVIDSON, FRANCES F. LOMBARD,<sup>1</sup> AND RAY R. HIRT<sup>2, 3</sup>

(WITH 4 FIGURES)

In a survey of decay in boats (5), started in 1941, an attempt was made to isolate and identify the fungus involved in each case investigated. Decay fungi were obtained from about one-third of the samples from which isolations were attempted. In many instances the fungus was inactive in the selected wood sample or molds had invaded the decayed wood so completely that the fungus causing the damage could not be isolated. In some samples the fungus obtained may not have been the cause of the principal damage but in general those fungi recovered most frequently are believed to be among the more important species causing decay. Certain fungi, such as *Poria incrassata* (Berk. & Curt.) Burt, which die rather quickly on drying (10), probably were involved in some cases of extensive damage although they were not identified during this limited survey. A more exhaustive study would result in obtaining additional species.

### TYPES OF ROT ENCOUNTERED

Most of the decays in wooden boats, as in buildings, are of the brown carbonizing type. The ratio is different from that usually encountered in living or dead trees in the forest where white and brown rots occur in about the same proportion.

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<sup>2</sup> Professor, Department of Forest Botany and Pathology, The New York State College of Forestry, Syracuse, New York.

<sup>3</sup> The authors are indebted to many individuals for collecting and sending in samples of decay.

During the present survey there were no important white rots found in softwood boat timbers but some were encountered in those of hardwoods. *Polyporus versicolor* Fr. was isolated from mahogany sapwood twice and was found fruiting on another specimen. *P. pargamensis* Fr. was isolated once from beech. Both of these fungi were present mainly because non-durable wood had been used. The white pocket rot, caused by *Stereum frustulosum* Fr., was the only important boat decay that would be placed in this group. It is one of the most important oak heart rots (4) and its common presence in oak boat timbers may have been due in part to the use of wood already infected.

The numerous brown rots are very much alike in appearance and usually they cannot be identified except from associated fruiting bodies or by studying the isolated organisms in pure culture. A number of the brown rots (Table 1) were obtained from both hardwoods and softwoods, notably those caused by *Poria microspora* Overh. and *P. xantha* (Fr. ex Lind) Cke. Although many fungi are known to invade both hardwoods and softwoods, some dual invasions were undoubtedly caused by the fact that in boat construction both types of wood are commonly used in close association.

Table 1 indicates the type of rot and kind of wood involved in each decayed specimen from which a fungus was obtained.

#### FUNGI ISOLATED FROM BOAT DECAY

It early became apparent that identification, by means of cultures, of most of the important species would be difficult. The initial difficulty was due in a large part to the fact that the most frequently isolated species, *Poria microspora* (FIG. 1, G to I), had not been described at that time. Cultures that bore the name *Trametes serialis* Fr. and were similar to the boat-decay fungus were not regarded as properly belonging in that species. Spore cultures from a poroid fruiting body proved to be the same as this common fungus but the sporophore was not identifiable. L. O. Overholts suggested that it might be the same as a fungus being studied by Miss M. K. Nobles and an exchange of cultures showed that the fungi

TABLE 1  
DATA ON FUNGI ISOLATED FROM DECAY IN WOODEN BOATS

Fungi	Type of Decay	Number of Specimens	Kind of Wood	Part of Boat Affected		
				Hull	Deck	Others <sup>1</sup>
<i>Poria microspora</i> Overh.	Brown	23	Fir <sup>2</sup> 10, pine 5, oak 5, cypress 2, white cedar 1	11		5
<i>Poria xantha</i> (Fr. ex Lind) Cke.	do.	21	Pine 8, fir 6, oak 5, spruce 1, mahogany 1	5	5	6
<i>Poria oleracea</i> Davidson & Lombard	do.	12	Oak 11, mahogany 1	2		9
<i>Daedalea quercina</i> L. ex Fr.	do.	9	Oak 6, mahogany 1, pine 1, other 1	3	2	2
<i>Lenzites saepiaria</i> Wulf. ex Fr.	do.	7	Pine 6, fir 1	1	5	1
<i>Poria carbonica</i> Overh.	do.	7	Fir 5, other 2	2	1	1
<i>Stereum frustulosum</i> Fr.	White pocket	7	Oak 7	1	1	5
<i>Lenzites trabea</i> Fr.	Brown	4	Pine 2, Mexican mahogany 1, fir 1		2	2
<i>Fomes roseus</i> (Fr.) Cke.	do.	2	White cedar 2	2		
<i>Lenzites lepidus</i> Fr.	do.	2	Pine 2	1	1	
<i>Polyporus versicolor</i> Fr.	White	3	Mahogany (sapwood) 2, elm 1	1	1	1
Unidentified fungus (producing strong odor)	Brown	2	Fir 1, oak 1	2		
<i>Coniophora cerebella</i> Pers. (C. puteana (Schum. ex Fr.) Karst.)	do.	1	Oak 1			1
<i>Hydnum erinaceus</i> Fr.	White	1	Beech 1			1
<i>Merulius tremellosus</i> Fr.	do.	1	Oak 1			1
<i>Polyporus pargamensis</i> Fr.	White	1	Beech 1	1		
<i>Polyporus sulphureus</i> Fr.	Brown	1	Oak 1			1
<i>Polyporus palustris</i> Berk. & Curt.	do.	1	Pine 1		1	
<i>Poria nigra</i> Berk.	do.	1	Oak 1		1	
<i>Trametes serialis</i> Fr.	do.	1	Fir 1			1
Miscellaneous unidentified fungi	Undetermined	8	Pine 3, mahogany 2, fir 1, undetermined 2		1	6

<sup>1</sup> Mostly chines, hatch coverings, fenders, and masts.<sup>2</sup> Fir as used here is mostly Douglas-fir.

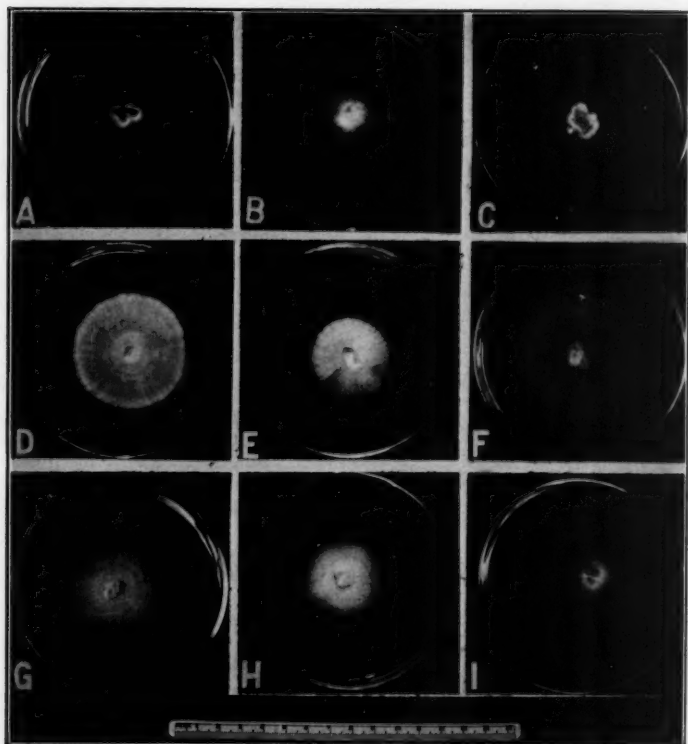


FIG. 1. One-week-old cultures of boat decay fungi grown on 2 per cent malt agar at room temperature. A, *Fomes roseus*; B, *Lentinus lepideus*; C, *Lenzites saepiarum*; D, *Lenzites trabea*; E, *Poria oleracea*; F, unidentified, odoriferous fungus; G to I, *Poria microspora*; G, isolate No. 106 from Douglas-fir decay in yacht "America"; H, Madison isolate No. 575; I, isolate from Douglas-fir log in Oregon.

were the same. It was described as *Poria microspora* Overh. in 1943 (6).<sup>4</sup>

<sup>4</sup> In a paper that appeared since this manuscript was written, L. O. Overholts (Mycologia 38: 674-675, 1946) presents evidence to show that his *Poria microspora* is a synonym of *Poria monticola* Murrill. A part of the type specimen of *P. monticola* (Weir No. 61) in the Bureau of Plant Industry herbarium at Beltsville, Maryland, appears to be the same as *Poria microspora* and is the best specimen of this fungus we have seen.

At the same time that *Poria microspora* was being studied, another species was causing similar confusion. The cultures seemed quite similar to those of *Fomes officinalis* Vill. ex Fr., but in several instances fragments of poroid fruiting bodies were attached to the decayed wood. These cultures when compared with available *Poria* cultures were identified as the new species *Poria carbonica* Overh. (6).

The species *Poria xantha* was occasionally found fruiting in typical condition but cultures of it are so variable that classification on the basis of cultures alone was sometimes difficult. The mycelium in some of the cultures had very little color, and in others developed considerably more slowly than in the typical ones.

Most of the species listed in Table 1 also occur under forest conditions. Those that are primarily oak-decay species, such as *Poria* sp. (herein described as *Poria oleracea* sp. nov.), *Dacdalea quercina* L. ex Fr., and *Stereum frustulosum*, are known to cause heart rots (4). Those that occur most frequently in softwood, such as *Poria microspora*, *P. xantha*, and *P. carbonica*, are not known to be important in living trees.

Those fungi which have been described previously from cultures (2, 4) and several of the more important ones studied by Nobles (6) have not been redescribed here. Besides occurring in living oak and in ship timbers, *P. oleracea* (previously described as *Poria* sp. (4)) was also isolated by Roth (9) from oak ties and undoubtedly is a fairly common fungus although it has not been possible to refer it to any of the previously known species. The methods employed in studying the cultures and the arrangement of the descriptions in this paper are similar to those given previously (4).

***Poria oleracea* Davidson and Lombard n. sp.<sup>5</sup> (FIG. 1, E; FIG. 2, A; FIG. 3)**

Pori in areas 3-6 mm. latas in ligno quercino in ampullis evolventes, delicatuli, dissepimentis tenuibus praediti, magnitudine formaque irregulariter, 2-3 per mm., 1-4 mm. longi, pallide alutacei usque vetusti et deteriores cervini, in siccitate contracti; basidia 9-13  $\times$  4-6  $\mu$ ; basidiosporae 5-7  $\times$  2-3  $\mu$ , hyalinae, aculeolatae; chlamydiosporae in substratu abundantes, late ovoideae, 5-10  $\times$  3-6  $\mu$ , leves, hyalinae.

<sup>5</sup> The Latin description was prepared by Edith K. Cash, Associate Mycologist, Division of Mycology and Disease Survey, Bureau of Plant Industry, Soils, and Agricultural Engineering, U. S. Department of Agriculture.

Pores developing in small patches (3-6 mm. or more across) on oak wood in flasks, delicate and thin-walled, irregular in size and shape, 2-3 per mm., 1-4 mm. long, "light buff" <sup>o</sup> to "tawny" when

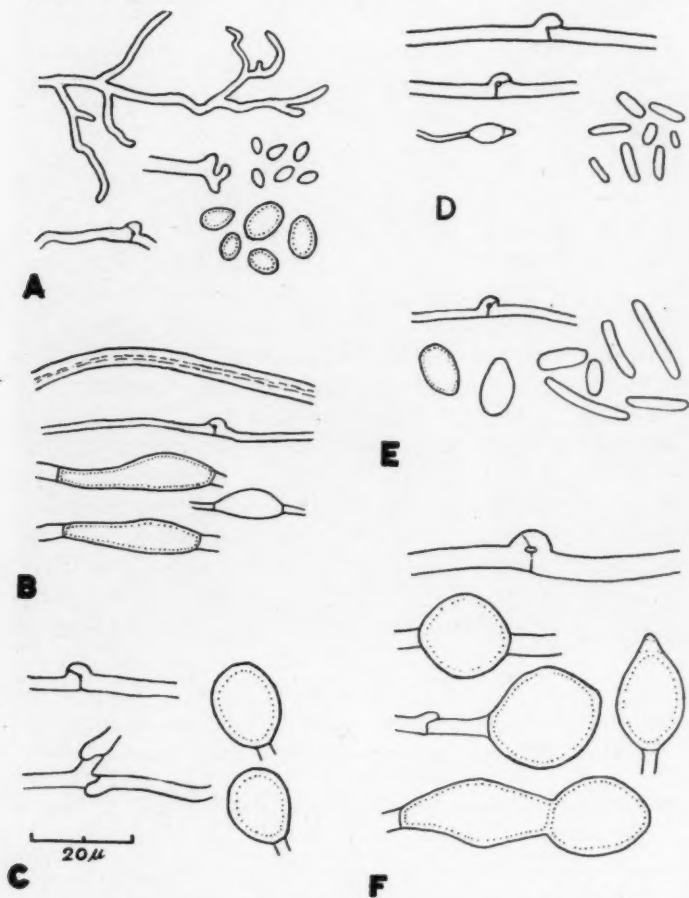


FIG. 2. *A*, *Poria oleracea*: hyphae, chlamydospores, and basidiospores. *B*, *Fomes roseus*: hyphae and chlamydospores. *C*, *Lentinus lepideus*: hyphae and chlamydospores. *D*, *Lenzites saepiaria*: hyphae, thin-walled chlamydospore, and oidia. *E*, *Lenzites trabea*: hypha, thin-walled chlamydospores, and oidia. *F*, unidentified fungus: hypha and chlamydospores.

<sup>o</sup> Colors in quotation marks according to Ridgway (8).

old and deteriorating, shrinking considerably on drying; basidia  $9-13 \times 4-6 \mu$ ; basidiospores  $5-7 \times 2-3 \mu$ , hyaline, somewhat pointed at ends; chlamydospores abundant in substratum, broadly ovoid,  $5-10 \times 3-6 \mu$ , smooth-walled, hyaline.

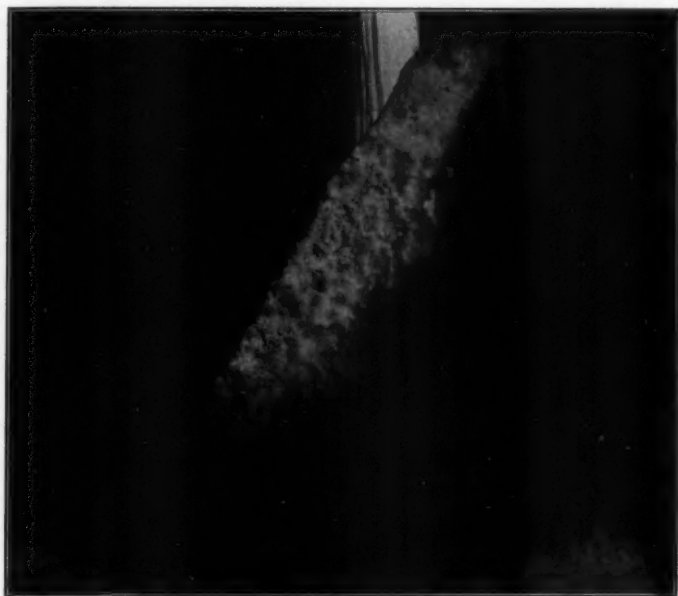


FIG. 3. Sporophore of *Poria oleracea* developed on sterilized oak wood in flask.

Type sporophores (F. P. No. 48282), developed on white oak wood in flasks from cultures isolated from rot in oak, deposited in the Mycological Collections of the Bureau of Plant Industry, United States Department of Agriculture.

Key pattern.<sup>7</sup>—A-O-M-1-2-5-6-10

<sup>7</sup> The key pattern has been explained previously (4, pages 9 to 13). The first letter represents color of mat (A, white; B, white, then yellow or brown; C, yellow, etc.). The second letter of the key pattern represents positive (P) or negative (O) oxidase reaction. The third letter indicates growth rate. The numbers indicate presence of certain microscopic features such as clamps, chlamydospores, conidia, etc. The key pattern is useful in arranging the fungi, based on their characteristics in culture.



*Growth characteristics.*—Growth medium, forming a mat 7 to 8.5 cm. diameter in fourteen days; mat short cottony to floccose cottony, usually zonate, and occasionally forming definite sectors; fragile, thin (agar showing through), white or rarely with a slightly yellowish area about the center; nodulose, waxy patches develop on the surface mycelium, at times abundant before fourteen days, but usually requiring a longer time to form, often producing basidia and basidiospores; margin proper white, floccose cottony, even; odor that of rotten cabbage in old cultures; negative oxidase reaction.\*

*Hyphal characteristics.*—Staining hyphae of two kinds, one 1–5  $\mu$  diameter, with simple clamps, much-branched, the other 4–8  $\mu$  diameter, smooth, little-branched, with simple and multiple clamps, becoming thick-walled, hyaline on aging, often with clublike ends where clamps have broken; chlamydospores 5–10  $\times$  3–6  $\mu$ , ellipsoid, with medium-thick hyaline walls, very abundant, in old cultures surface mycelium practically all turned into chlamydospores; basidia formed in the nodulose patches on surface sometimes fairly abundant in fourteen days.

*Temperature relations.*—Optimum approximately 30° C. Average mat diameters in seven days in the dark at constant temperatures follow: 2.4 cm., 20°; 3.9 cm., 25°; 5.4 cm., 30°; 4.8 cm., 35°; 0, 41° C.

*Test-tube cultures.*—In twenty-eight days mat appressed, fragile, pulverulent to floccose or somewhat nodulose, white, often with raised waxy patches on upper part of slant that develop into shallow pores; glass opposite slant coated with a feathery or cottony mycelium; poroid fruiting in small patches on upper part of agar slant and on glass opposite slant, at first meruloid, later definitely poroid with pores irregular in shape and size, mostly small (0.3 mm.) but with thick walls and remaining fairly shallow; "light buff" to "pinkish buff" in color with brown developing at margins where drying occurs; spore print white, often forming on side of test-tube opposite poroid areas; basidia oblong, 9–13  $\times$  4–6  $\mu$  with four sterigmata; basidiospores 5–7  $\times$  2–3  $\mu$ , elongate with somewhat pointed ends, hyaline.

\* Fungi causing brown carbonizing rot in wood usually give no reaction when grown on agar containing gallic or tannic acid whereas white rot fungi cause a dark color reaction when grown on such media (3).

*Type of decay.*—Brown rot in oak heartwood. Isolated twelve times from boat-decay specimens, one of which was from mahogany wood.

*Remarks.*—This species was isolated thirteen times from oak heart rot specimens (4) and several times from oak ties by Roth (9). It is somewhat similar to *Polyporus compactus* Overh. (4, 7) in the abundant chlamydospore development but is different in other respects. Sporophores develop readily on oak blocks in flask cultures, especially on the glass where the block touches the side of the flask.

FOMES ROSEUS (Fr.) Cke. (FIG. 1, A; FIG. 2, B)

*Key pattern.*—B-O-M-1-2-10

*Growth characteristics.*—Growth medium, in seven days forming a mat 3 to 4 cm. diameter; mat dense, appressed, sometimes with thicker areas radiating from the center, white, then "light pinkish cinnamon" over central area in ten to fourteen days to "cinnamon" on inoculum; marginal zone broad, thin, appressed to submerged, margin proper inconspicuous, fimbriate; odor similar to that of green apples (1); oxidase reaction usually negative but sometimes a dark zone develops in gallic acid medium under inoculum block but does not advance with the new growth.

*Hyphal characteristics.*—Both submerged and young superficial hyphae staining, 1.5–4  $\mu$  diameter, some superficial hyphae becoming thick-walled and non-staining, few groups of brown-walled hyphal segments in fourteen-day-old cultures; clamps abundant; chlamydospores few, not conspicuous, long and narrow, 18–25  $\times$  5–6  $\mu$ .

*Test-tube cultures.*—About the same as in Petri dishes with dense mat forming only over old inoculum, sometimes developing pores in three to four weeks.

*Type of decay.*—Brown carbonizing rot in Atlantic white cedar hull planking. Isolated twice.<sup>9</sup>

*Remarks.*—It is difficult to distinguish *Fomes roseus* from *F. subroseus* (Weir) Overh. but the latter is reported to be faster

<sup>9</sup> One of these was isolated by C. Audrey Richards and sent to the Division of Forest Pathology, Beltsville, Maryland, for identification.

growing than the former (1). Since the cultures isolated from boats were slower growing than those recorded for *F. subroseus*, they are here referred to *F. roseus*.

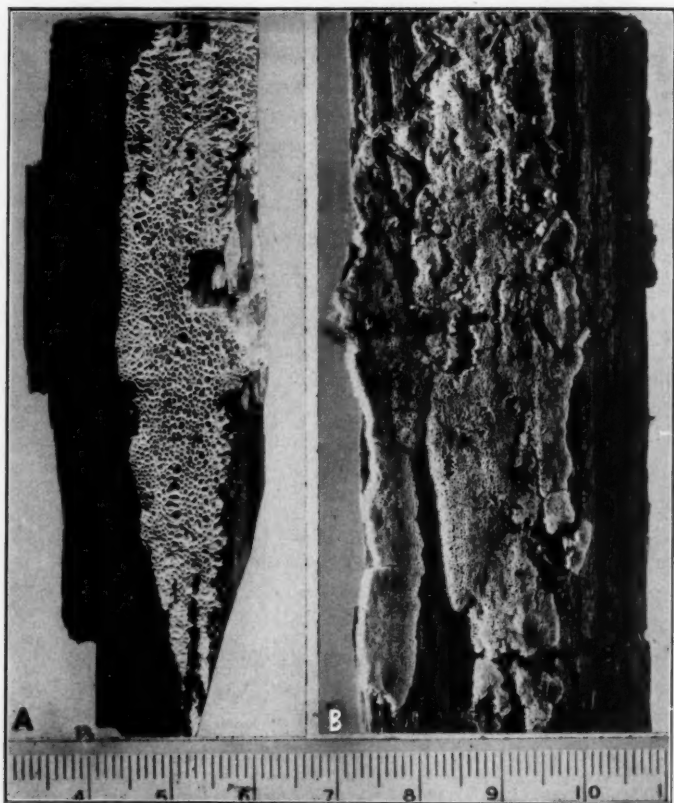


FIG. 4. *A*, sporophore of *Poria microspora* fruiting on pine wood from the whaler "Charles Morgan." *B*, fruiting of *Poria xantha* on Douglas-fir from a barge.

LENTINUS LEPIDEUS Fr. (FIG. 1, *B*; FIG. 2, *C*)

*Key pattern*.—A-O-I-1-2-10

*Growth characteristics*.—Growth moderately rapid, forming a mat 9 cm. diameter in fourteen days; mat tufted, woolly cottony,

raised, peeling off from substratum easily and breaking up readily, white with buff to brownish tints at fourteen days; negative oxidase reaction.

*Hyphal characteristics.*—Hyphae staining with eosin, variable in size, 1.8 to 6  $\mu$  diameter, occasionally branching; clamps numerous; chlamydospores abundant, almost round, large, 10–20  $\times$  9–13  $\mu$ , walls medium thick (about 1  $\mu$ ).

*Test-tube cultures.*—Forming a dense, buff to brown mat over slant, with lumps or short fingerlike stalks developing in about three weeks. Fertile sporophores have never been observed in culture but the short stalks usually start to develop.

*Type of decay.*—Brown carbonizing decay from pine boat timbers. Isolated twice from boats. This species has been isolated from heart rot in southern pines<sup>10</sup> and from ponderosa pines in California (11).

*Remarks.*—This species is easily identified in test-tube cultures by the numerous short stalks that develop.

LENZITES SAEPIARIA Wulf. ex Fr. (FIG. 1, C; FIG. 2, D)

*Key pattern.*—A-O-M-1-2-4-10

*Growth characteristics.*—In fourteen days forming a mat 5.0 cm. diameter; mat white, thin, appressed, cottony to powdery; marginal zone irregular, sometimes thin and inconspicuous to powdery; margin proper uneven, fimbriate; negative oxidase reaction.

*Hyphal characteristics.*—Both submerged and superficial hyphae staining with eosin, thin, 1.8 to 3.5  $\mu$  diameter, much-branched; oidia abundant in very young cultures, cylindrical or sometimes ovoid, 4–14  $\times$  2–3  $\mu$ , thin-walled; chlamydospores few, inconspicuous, 8–12  $\times$  5–7  $\mu$ , thin-walled.

*Test-tube cultures.*—Three- to four-week-old test-tube cultures white and powdery near margins of growth but brown ("pinkish buff" to "cinnamon") on older areas of mat, coloring substratum brown near upper part of slant; mat thin powdery with cottony tufts or patches, seldom if ever raised or filling tube above slant; abortive fruiting bodies forming occasionally.

<sup>10</sup> Unpublished data.

*Type of decay.*—Causes a brown carbonizing decay. Seven specimens collected from boats—mostly from pine deck planking containing some sapwood.

LENZITES TRABEA Fr. (FIG. 1, D; FIG. 2, E)

*Key pattern.*—B-O-I-1-2-4-10

*Growth characteristics.*—Growth moderately rapid, forming a mat 9 to 10 cm. diameter in fourteen days; mat fairly dense, floccose cottony to margin, "light buff" to "light ochraceous-buff" with darker "clay color" patches forming on some isolates, raised, about 2 mm. thick, somewhat tufted or uneven on the surface; odor sweet; negative oxidase reaction.

*Hyphal characteristics.*—Hyphae staining, thin-walled; submerged hyphae small, much-branched, 1.5 to 3  $\mu$ , with a few larger, up to 5  $\mu$  diameter; surface hyphae uniformly small, 2-4  $\mu$  diameter, few branches; clamps numerous on both submerged and superficial hyphae; oidia abundant in young cultures, segments of hyphae, 5-25  $\times$  2.5-3.5  $\mu$ , hyaline, thin-walled, cylindrical, sometimes elongate-ovoid.

*Test-tube cultures.*—Loose to dense plug of "light ochraceous-buff" mycelium grows up into neck of tube, sometimes growing into the cotton plug.

*Type of decay.*—Brown carbonizing decay, usually in pine sapwood. Isolated three times from pine and Mexican mahogany.

UNIDENTIFIED FUNGUS (*producing strong odor*) (FIG. 1, F; FIG. 2, F)

*Key pattern.*—A-O-I-1-2-10

*Growth characteristics.*—Growth fairly rapid, forming a mat 7.5 cm. diameter in seven days; mat raised, loose, feathery, sometimes collapsing in central zone after six days; odor very disagreeable (putrescent) at end of fourteen days; negative oxidase reaction, good growth (4.5 cm. in 7 days) on gallic acid medium and poor growth on tannic acid but fluffing up in coarse strands and growing over the underside of Petri dish lid.

*Hyphal characteristics.*—Submerged and superficial hyphae staining with eosin, much-branched, small to medium sized, few

large diameter hyphae with clublike ends where broken at clamps; clamps numerous and conspicuous on small and large hyphae; chlamydospores abundant, large, ovoid to almost round,  $10-16 \times 8-12 \mu$ , thick-walled, intercalary or terminal, sometimes in short chains.

*Test-tube cultures.*—Loose feathery mat about same as in Petri dishes, dull white with no color on aging. Disagreeable odor at two to four weeks.

*Type of decay.*—This fungus has not been proved to cause decay in wood but it was isolated from brown rot in coniferous wood. The negative oxidase reaction also indicates that it is a brown rot fungus. It was isolated twice.

#### FUNGI FRUITING ON BOATS

Some of the boat decay fungi frequently develop small somewhat abortive sporophores so that, without a thorough knowledge of the species, identification is difficult. Well-developed fresh sporophores of *P. xantha* (FIG. 4, B) have a characteristic yellow color and cheesy texture but color and texture are not so characteristic after specimens have aged. Prominent hyphal strands are also formed by this species and are useful as an aid to identification.

Most strains of *Poria microspora* fruit readily in culture but only one identifiable sporophore was collected during the survey (FIG. 4, A). The sporophores are delicate and soon disintegrate when formed under natural conditions.

*Lenzites saepiaria* fruits readily on deck planking and was obtained as sporophore specimens more frequently than isolated from decayed wood. Most of the sporophores were from pine deck planking of barges—probably where some sapwood was present.

As mentioned previously, somewhat abortive poroid fruiting bodies were present on two of the specimens from which *Poria carbonica* was isolated. Such fruiting is difficult to identify without the aid of cultures. Table 2 lists the sporophores that were collected. Several of these fungi found fruiting but not isolated may be important decay fungi, especially *Paxillus panuoides*, which is known to cause a brown rot in buildings.

TABLE 2  
FUNGI FRUITING ON DECAYED WOOD IN BOATS

Species of Fungi	Number of Times Collected	Type of Boat	Places Collected	Collectors
<i>Poria xantha</i> (Fr. ex Lind) Cke.	9	Barge 5, cruiser 3, schooner 1	Norfolk, Va. 3, Brewerton, N. Y. 3, Oakland, Calif. 1, Tacoma, Wash. 1, Noank, Conn. 1	Davidson 3, Hirt 3, Kimney 1, Englerth 1, Boyce 1
<i>Lenzites saepiaria</i> Wulf. ex Fr.	6	Scow 1, barge 4, cruiser 1	New Orleans, La. 3, Erie, Pa. 1, Kingston, Ont. 1, Norfolk, Va. 1	Verrall 3, Hirt 2, Davidson 1
<i>Poria carbonica</i> Overh.	2	Scow 1, unknown 1	Seattle, Wash. 1, Kingston, Ont. 1	Englerth 1, Hirt 1
<i>Stereum frutulosum</i> Fr.	2	Tug 1, barge 1	Curtis Bay, Md. 1, Kingston, Ont. 1	Davidson 1, Hirt 1
<i>Dacrydia quercina</i> L. ex Fr.	1	Barge 1	Kingston, Ont. 1	Hirt 1
<i>Omphalia campanella</i> (Fr.) Quél.	1	Scow 1	Kingston, Ont. 1	Hirt 1
<i>Paxillus panuoides</i> Fr.	1	Scow 1	Kingston, Ont. 1	Hirt 1
<i>Polyporus versicolor</i> Fr.	1	Barge 1	Kingston, Ont. 1	Hirt 1
<i>Poria microspora</i> Overh.	1	Whaler 1	Mystic, Conn. 1	Hansbrough and Spaulding 1
<i>Peniophora</i> sp. <sup>1</sup>	1	Scow 1	Kingston, Ont. 1	Hirt 1
<i>Poria</i> sp. <sup>1</sup>	4	Scow 1, barge 2, tug 1	Kingston, Ont. 2, Tacoma, Wash. 1, New Orleans, La. 1	Hirt 2, Englerth 1, Verrall 1
<i>Schizophyllum commune</i> Fr.	1	Cabin cruiser 1	Brewerton, N. Y. 1	Hirt 1
<i>Stereum</i> sp. <sup>1</sup>	1	Cruiser 1	Brewerton, N. Y. 1	Hirt 1
<i>Xylaria</i> sp. <sup>1</sup>	1	Barge 1	Kingston, Ont. 1	Hirt 1

<sup>1</sup> Fruiting bodies collected but fungi not isolated from decay; importance uncertain.



## SUMMARY

Twenty species of wood rot fungi were isolated from or collected on specimens of decayed wood in boats, barges, tugs, schooners, motor boats, etc., and sporophores of several additional species known to cause decay in wood were collected. The important causes of softwood decay were *Poria microspora*, isolated 23 times; *P. xantha*, 21 times; *P. carbonica*, 7 times; and *Lenzites saepiaria*, 7 times. The more important hardwood fungi were *Poria oleracea*, obtained 12 times; *Daedalea quercina*, 9 times; and *Stereum frustulosum*, 7 times.

The more important hardwood fungi were species that also occur as heart rots in living oaks. *Poria oleracea* had previously been isolated from heart rot in oak and is described in this paper as a new species. The more important species in softwood boat timbers are known to be important in decay of buildings and other wood products, but are not known to be important causes of heart rot.

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## PEZICULA MORTHIERI ON RHAMNUS<sup>1</sup>

J. WALTON GROVES<sup>2</sup>

(WITH 3 FIGURES)

The genera *Dermea* and *Pezicula* are of special interest because of the various types of conidial stages occurring in the different species. It has been shown by Groves (1939, 1940, 1946) that the form of the conidial fruiting body may vary greatly in both genera, but that in most species of *Dermea* the conidia are elongate to subfiliform whereas in *Pezicula* they are oblong-ellipsoid to ovoid. Two exceptions to this general rule have been noted, *D. acerina* (Pk.) Rehm (Groves 1938) and *P. alnicola* Groves (Groves 1940). It was stated by Groves (1946) that a third exception was known in a species occurring on *Rhamnus* and known as *Dermea micula* (Fr.) Rehm, and that a more detailed account would be published later.

The specific name used by Rehm (1889) was, however, based originally on the conidial stage and since the perfect stage was described earlier by Fuckel (1870) as *Cenangium Morthieri* the latter name must be considered valid according to the International Rules of Nomenclature.

As far as I know, the apothecia of this species have not been reported in North America, but in 1936 Mr. G. Lapointe collected it on *Rhamnus alnifolia* at St. Alphonse de Caplan, Bonaventure Co., Quebec, and I received fresh specimens in excellent condition from which I was able to obtain cultures from both ascospores and conidia.

Fuckel's original description was apparently based on the specimen distributed in Fungi Rhenani 2278. The specimen under this number in the Farlow Herbarium, Harvard University, was examined and it agreed with the Canadian collection. In Fuckel's

<sup>1</sup> Contribution No. 890 from the Division of Botany and Plant Pathology, Science Service, Department of Agriculture, Ottawa, Canada.

<sup>2</sup> Plant Pathologist, Central Laboratory, Ottawa.

account, he described and figured four spored asci, but in the Canadian material and in the specimen of Fung. Rhen. 2278 that I examined, the asci were eight spored (FIG. 3). It is possible that Fuckel's material may have been a mixture of this species and *Pezicula Frangulae* (Pers.) Fckl., which has four spored asci and also occurs on *Rhamnus*, and that he may have examined a mount of the latter.

Fuckel noted its association with the conidial stage which he knew as *Micula Mougeotii* Duby and suggested the genetic connection, but this relationship was not established by cultural methods. The pycnidia are very striking in appearance with their long, snow-white beaks, and are much more conspicuous than the apothecia (FIG. 1). Fuckel stated that he had always thought this was the conidial stage of *Tympanis* (*Pezicula*) *Frangulae* until he saw this collection and observed it actually arising on the same "raschen." The conidial stage of *Pezicula Frangulae* has since been established by Wollenweber (1939) as *Cryptosporiopsis versiformis* (A. & S.) Wollenw.

Von Höhnelt (1912) pointed out that the structure of the conidial stage had been misinterpreted by Saccardo (1879) and that he had placed it erroneously in the Stilbaceae as *Atractium Therryanum*. It was again described accurately by Dearness and House (1925) as *Sphaerographium niveum*, based on North American material, but presumably they did not have the perfect stage.

The generic position of this fungus is open to question. In color and consistency of the apothecia it is closer to *Pezicula* than to *Dermea*, but it has the narrow ascospores and asci that are more typical of *Dermea* than *Pezicula* and the conidia are definitely closer to the *Micropera* type than the *Cryptosporiopsis* type. Nannfeldt (1936) stated that it was a *Dermea*. It is one of those border-line species which could be placed in either genus with about equal justification. Because of the color and consistency of the apothecia I prefer to regard it as a somewhat aberrant *Pezicula*. The following description is based on the Canadian material.

***Pezicula Morthieri* (Fuckel) n. comb. (FIGS. 1-3)**

*Cenangium Morthieri* Fuckel. Symb. Myc. p. 272. 1870.

*Cenangella Morthieri* Sacc. Syll. Fung. 8: 592. 1889.

*Phaeangella Morthieri* Sacc. & D. Sacc. Syll. Fung. 18: 128. 1906.

*Dermatea micula* Rehm. Rab. Kr. Fl. I, 3: 261. 1889. St. conid.

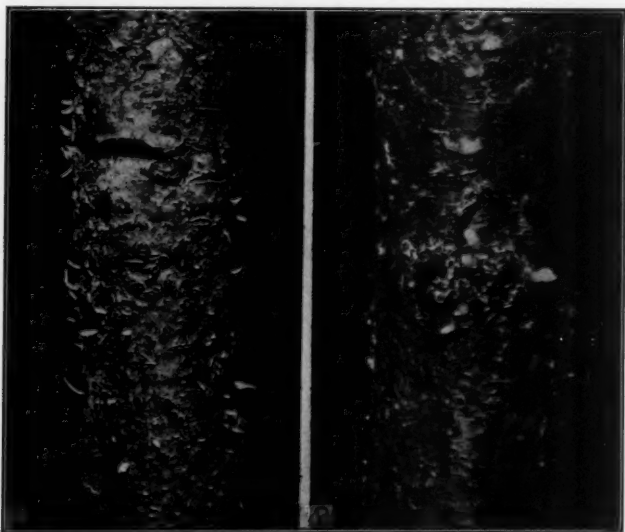


FIG. 1. Apothecia and pycnidia of *Pezicula Morthieri* from the Quebec specimen.

FIG. 2. Pycnidia produced on a twig of *Rhamnus* in culture originating from ascospores.  $M = 4\times$  approx.

*Sphaeria micula* Fries. Elench. Fung. 2: 101. 1828.

*Micula Mougeotii* Duby. Kl. Herb. Myc. 636. 1844.

*Atractium Therryanum* Sacc. Mich. 1: 535. 1879.

*Sphaerographium niveum* Dearn. & House. Bull. N. Y. St. Mus. 266: 89. 1925.

Apothecia erumpent, scattered, separate or occasionally two or three in a cluster, circular or slightly irregular, sessile, narrowed below, 0.4–0.8 mm. in diameter, 0.2–0.5 mm. in height, yellowish brown to reddish brown, brittle, waxy, more fleshy when moist; hymenium at first concave, becoming plane to convex, darker than the excipulum, dark brown to almost black, becoming brighter when moist; tissue of the hypothecium compact, pseudoparenchy-

matous, composed of hyaline, usually slightly elongated cells  $5-12\ \mu$  in diameter with the walls thickened and gelatinized, arranged in vertically parallel rows, curving obliquely toward the outside, walls of the outer cells yellowish; subhymenium indistinct; asci cylindric-clavate, short stalked, eight spored,  $(60)-65-95-(110) \times (7.5)-8-11-(14)\ \mu$ ; ascospores ellipsoid-fusiform, hyaline or pale yellowish, straight or slightly curved, one to four celled, irregularly biseriate,  $(12)-15-20-(24) \times (3.5)-4-6-(7)\ \mu$ ; paraphyses hyaline, filiform, septate, simple or branched,  $1.5-3.5\ \mu$  in diameter, the tips not at all or very slightly swollen, and forming a slight epithecium.

Conidial fruiting bodies erumpent, scattered, separate or occasionally two or three together, sometimes arising from the same basal stroma as the apothecia, cylindric-conic, tip truncate,  $200-400\ \mu$  in diameter at the base,  $70-150\ \mu$  in diameter at the tip,  $0.5-1.0$  mm. in height, usually snow-white, sometimes darker, brittle, more fleshy when moist; tissue of the basal stroma compact, pseudoparenchymatous, composed of hyaline, almost isodiametric cells  $5-10\ \mu$  in diameter with the walls somewhat gelatinized, tissue of the beak composed of two zones, an outer pseudoparenchymatous zone similar to the basal stroma, and an inner zone composed of hyaline, parallel, thick-walled hyphae  $3-5\ \mu$  in diameter; conidiphores hyaline, cylindric, septate, branched,  $25-40 \times 2.0-2.5\ \mu$ ; conidia hyaline, filiform, slightly curved to almost straight, pointed at the ends, one to four celled,  $(30)-45-65-(70) \times (2.0)-3-4-(5.0)\ \mu$ .

HOST: *Rhamnus alnifolia* L'Her.

EXSICCATI: Fuckel Fung. Rhen. 2278 (*Cenangium Morthieri* Fekl. Type); 1763 (*Micula Mougeotii* Duby); Roum. Fung. Sel. Gall. Exs. 367 (*Sphaeria Micula* Fr.); Rehm Ascom. 1411 (*Dermatea Micula* (Fr.) Rehm).

SPECIMENS EXAMINED: Herbarium of the Division of Botany and Plant Pathology, Science Service, Ottawa, Canada 19242, on *Rhamnus alnifolia*, L'Her., St. Alphonse de Caplan, Bonaventure Co., Que. (JWG-499).

The cultures from ascospores were similar in all respects to those obtained from the conidia and both ascospore and conidial cultures produced conidial fructifications in culture. On malt extract agar the cultures grow rather slowly, reaching a diameter of  $3.5-4.0$  cm. in three weeks. The colonies are circular to slightly lobed with the margin even to slightly fimbriate, whitish to pale buff, aerial mycelium short, velvety-tomentose, slightly zoned, surface flat. The conidial fructifications are fleshy, at first globose,

become irregular in shape, rarely form a beak, usually tear open irregularly at the top and the spores emerge in whitish masses.

On sterilized twigs of *Rhamnus* very little aerial mycelium was produced except for a few whitish tufts around the point of inoculation. The conidial fruiting bodies (FIG. 2) were erumpent, scat-

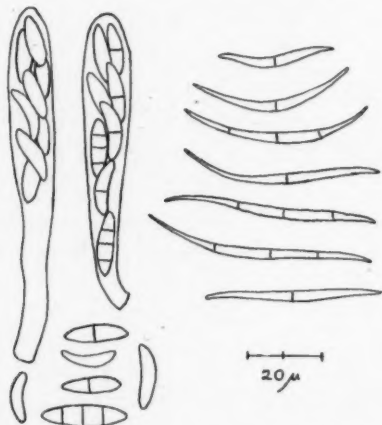


FIG. 3. *Pezicula Morthieri*; asci, ascospores and conidia.

tered, not very abundant, at first rounded, and fleshy. They sometimes split open with the spores emerging, but sometimes develop a conical, white beak as in nature, 0.3–0.8 mm. in diameter and 0.8–1.5 mm. in height, with the conidia similar to those found in nature.

#### ACKNOWLEDGMENTS

I am indebted to Mr. I. L. Conners who generously placed the Quebec collection at my disposal for cultural studies, and to the late Dr. D. H. Linder for permission to examine the type material in Fungi Rhenani.

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## BRIEF NOTES ON THE GENERA STEREOSTRATUM MAGN. AND ANTHOMYCE-TELLA SYD.

M. J. THIRUMALACHAR

(WITH 7 FIGURES)

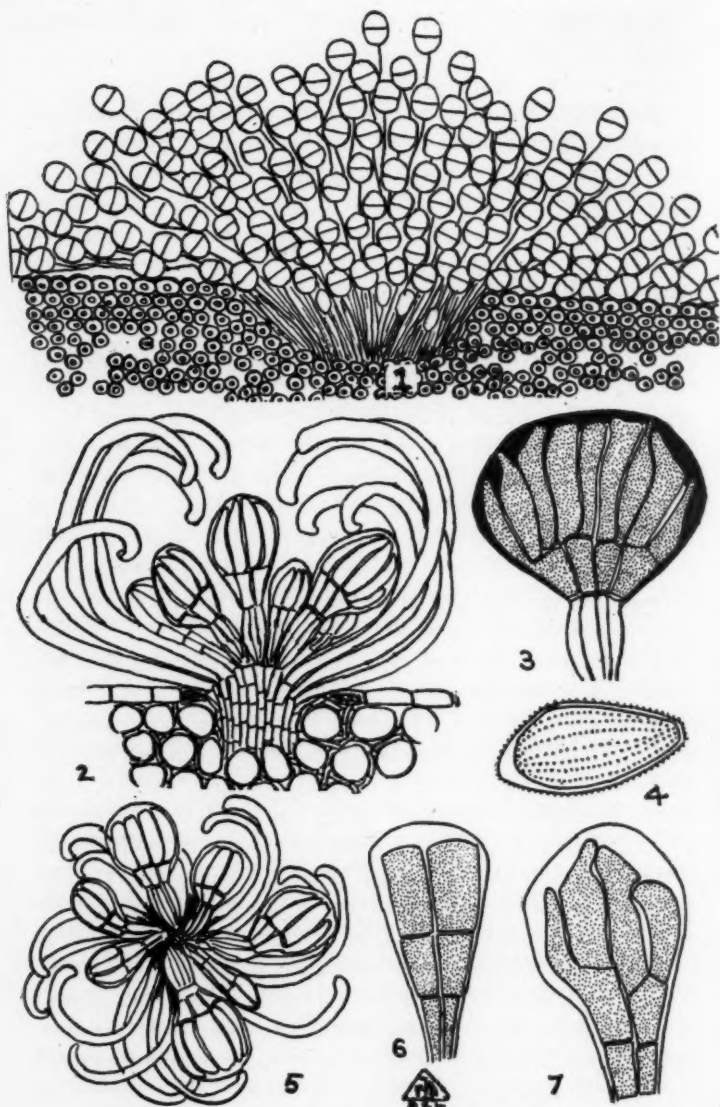
The bamboo rust *Stereostroma corticioides* (Berk. & Br.) Magn. belongs in a monotypic genus established by Magnus in 1899 from material collected in Japan. The rust was originally described by Berkeley and Broome (1877) as *Puccinia corticioides* and later as *Puccinia Schottmullerii* by Hennings (1893). The presence of two-celled hyaline teliospores grouped in sori which form large *Corticium*-like crusts measuring sometimes up to 10 cm. in diam. and each spore with three germ pores in each cell were considered by Magnus as diagnostic characters for the genus *Stereostroma*. The genus has been recognized by Dietel (1928) and others though the Sydows (1904) in the *Monographia Uredinearum*, I, treated it as a synonym of *Puccinia* by retaining the name *Puccinia corticioides* Berk. & Br. However, in Sydow's *Fungi Exotici Exsiccati* No. 953, collected in Kyoto, Japan, in 1933 by Kiyoshi Aoki, the rust has been distributed under the name *Stereostroma corticioides*. A small fragment of this specimen bearing the number 953 was made available for study through the kindness of Dr. Th. Arwidsson of the Natural History Museum, Stockholm. Although the type of the genus is recorded on *Bambusa metake* (= *Arundinaria japonica*), the collection made by Kiyoshi Aoki and distributed in Sydow's *Fungi Exotici Exsiccati* No. 953 is on *Phyllostachys bambusoides*.

Only uredia and telia are known for the genus. The uredia form striate sori developing urediospores. The telia which are present on the woody culms are of extreme interest because they form sori which are described as being *Corticium*-like and measure up to 10 cm. in diameter. Such large sized sori are very peculiar and so far unknown among other rust genera. In the material

at the disposal of the writer the telia had formed crusts measuring from 5 to 8 mm. in diam. and a study of the structure of the sorus and the development of the teliospores was made.

Sectioning of the material is rendered very difficult because of the sclerotic nature of the woody culms. However, thin sections were secured after softening the material. Observations indicate that the sorus is subepidermal, and formed by the displacement of the sclerotic cells of the host by hyphal strands. In transverse sections, the sorus appears as a concave depression lined with hyphal strands. From the base of the hymenium, a large number of pedicellate teliospores, which are two-celled and hyaline, are produced in succession. The teliospores so developed are not persistent but soon become separated from the sorus and are pushed upwards by the developing younger spores from the base. The younger teliospores are developed between two older ones and appear wedged in between them. The teliospores are therefore formed in succession from the base in the same manner as in the tribe Puccinosirae of Dietel (1928) and in particular as in the genus *Kernia* recently described by the writer (1946). In *Kernia*, however, the teliospores are grouped compactly to form long, semipermanent *Cronartium*-like spore tendrils. In *Stereostratum* on the other hand, the older teliospores are pushed laterally without any compact arrangement (FIG. 1) and the long pedicels, which are up to  $350\ \mu$  long, intertwine with one another, with the result that a spreading pulvinate cushion-like sorus is produced. As more spores are added from the base the upper spores are pushed farther off with the result that the diameter of the sorus increases. It has also been noticed that along the border of the sorus more and more basal cells are differentiated abstricting more spores, thereby widening the breadth of the basal hymenium. The development of such large sori is made possible by the method of spore formation mentioned above.

Mature teliospores are thin-walled, hyaline, two-celled, slightly or not at all constricted at the septa, not thickened at the apex, and measure  $24-34 \times 18-25\ \mu$ . The episporium is smooth to rugose, and shows three distinct germ pores. The pedicels are hyaline, intertwining with one another, persistent, up to  $350\ \mu$  long, and taper into thin hyphoid strands for the most part at the base.

FIGS. 1-7. *Stereostroma* and *Anthomycetella*.

The genus *Stereostratum* is placed in the Uropyxideae by Dietel (1928) who placed it in that tribe with some reserve, and considered it doubtful as to whether the mere presence of three germ pores would warrant its inclusion in that tribe. The other genera included under Uropyxideae all possess three distinct wall layers in addition to the number of germ pores, a character not duplicated in *Stereostratum*. The presence of more than one germ pore is known in a number of other genera not included under Uropyxideae and taking into consideration the development of teliospores in succession from the base of the hymenium it seems reasonable to place the genus *Stereostratum* in the tribe Pucciniosirae.

The genus *Anthomycetella* Sydow (1916) was established to accommodate a leaf rust on *Canarium villosum* from the Philippines, and was later described by Saccardo (1917) under the name *Reyesella* Sacc. The rust is characterized by the possession of telia having multicellular telial heads borne on compound pedicels composed of three to four hyphal strands. The teliospores are stated to be arranged in two tiers, the upper one having always a larger number of spores than the lower tier. The sori are covered by incurved cylindric paraphyses, which according to Sydow are coalescent at the base. The genus *Anthomycetella* is described as being closely related to *Anthomyces* Diet. but differing from it in the possession of a compound pedicel and lack of cysts, which, however, are characteristic of the latter genus. In the arrangement of the superposed teliospores, Sydow finds some relationship between the genus *Anthomycetella* and *Pleoravenelia*, but in the structure of the telial head the two show considerable difference.

A small fragment of an authentic specimen of *Anthomycetella Canarii* Syd., which is the only species known for the genus, was made available through the kindness of Dr. Th. Arwidsson. The material was collected in the type locality Los Baños, Philippines, by Baker, the identification being made by Sydow. A brief study of the material revealed certain interesting features not recorded in the descriptions, and it is considered worthwhile to place them on record.

Sections through the leaf indicated that prior to the formation of the telium, which is the only spore form so far known for the genus, strands of hyphae become grouped beneath the stoma and emerge by pushing apart the guard cells. The strands of hyphae bear the telial sorus above the epidermis with the pedicellate telial heads arising in the middle and the strongly incurved cylindric paraphyses developing along the margin (FIG. 2). The paraphyses possess a few septations here and there and completely envelope the telia. In the method of telial development there is very close resemblance with that of the uredia of *Crossospora Zizyphi* Syd. recently described by Mundkur and Thirumalachar (1946). When the sorus is scraped with a scalpel to get some spore mounts, the entire sorus gets detached by the abscission of the hyphal strands above the stoma. Consequently the paraphyses which are marginal and are attached to the hyphal strands appear to have coalesced at the base (FIG. 5).

The developmental stages of the telial heads have been followed as far as the scanty material at the disposal of the writer permitted. In the early stages of development, the sorus is composed of two to three layers of hyphal strands composing the pedicel and the same number of hyphal cells above them. From each of these hyphal cells, which by now become enclosed within a membrane, a teliospore is formed above, which elongates and becomes cylindric (FIG. 6). The same basal hyphal cell continues to abstrict off more teliospores laterally with the result that an umbellate cluster of teliospores is produced of which the centrally situated ones are the oldest and the marginal ones are youngest (FIGS. 3 and 7). Various stages in the formation of teliospores from the basal cell, which should more properly be considered a sporogenous basal cell, have been followed. All the spores thus produced are enclosed within a brownish-yellow wall which appears to become slightly gelatinous in later stages (FIG. 3). Consequently in a mature telial head we can observe the compound pedicel composed of three to four hyphal strands, three or four basal cells bearing 18 to 24 clavate to rectangular teliospores. These basal cells have been treated as teliospores by Sydow (1916), Dietel (1928) and others and consequently the telial heads are

described as having two superposed tiers of spores on a compound pedicel. The method of spore formation from each basal cell as well as a few cases of germination observed in some of the teliospores indicate beyond a doubt that the lower tier of cells are not of the nature of spores. In some of the mature telial heads that were noticed, the apex of the teliospores had prolonged into a four-celled promycelium. The germ tube breaks through the gelatinous envelope and the contents of the teliospore migrate into the promycelium thus formed. None of the lower tiers of cells had developed any such germ tubes, indicating that they are only of the nature of basal cells and not spores. In the figure of *Anthomycetella Canarii* given by Dietel (1928), two tiers of teliospores superposed over one another without any common membrane enclosing the telial heads are shown. A careful examination of the teliospores in the authentic specimen of *Anthomycetella Canarii* indicates that the illustration is not appropriate.

A few urediospores were seen intermixed with young telia, and separate uredia have not been observed in the material at the disposal of the writer. The uredia are so far unknown for the genus, only the telial stages having been described. The urediospores (FIG. 4) are ovate ellipsoid, thin-walled, hyaline in the exsiccated material, densely echinulate and without any distinct germ pores. The spores measure  $32-35 \times 16-20 \mu$ . The echinulations on the spore are arranged in definite vertical rows giving a striate appearance.

The genus *Anthomycetella* is placed in the tribe *Raveneliae* by Dietel on account of the formation of telial heads and the presence of compound pedicels. There is only umbellate cluster of spores produced from the basal sporogenous cells which are up to three to four in number and not two superposed tiers of spores as originally described. If we could imagine clusters of teliospores of a *Chaconia* like *C. Butleri* (Syd.) Mains as being enclosed within a common spore membrane to form a head, the whole telial head being borne on strands of hyphae, we arrive at the condition in the telial head of *Anthomycetella*.

In conclusion the writer wishes to acknowledge his grateful thanks to Dr. Th. Arwidsson of the Cryptogamic Herbarium,

Naturhistorika Riksmuseet, Stockholm, Sweden, for sending the valuable specimens used in the present study.

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#### DESCRIPTION OF FIGURES

- FIG. 1. Section through the telium of *Stereostroma corticioides*  $\times 150$ .  
FIG. 2. Section through the telium of *Anthomycetella Canarii*  $\times 200$ . FIG. 3. Mature telial head  $\times 400$ . FIG. 4. Urediospore  $\times 1000$ . FIG. 5. Surface view of the entire sorus from the scraped material  $\times 200$ . FIGS. 6 and 7. Development of teliospores from the basal cells  $\times 800$ .



## SELENOPHOMA LINICOLA SP. NOV. ON FLAX IN SASKATCHEWAN<sup>1</sup>

T. C. VANTERPOOL<sup>2</sup>

(WITH 10 FIGURES)

At harvest time in 1944 a fungus with mature pycnidia and belonging to the genus *Selenophoma* Maire (5 and 7) was collected on the fine branches and pedicels (FIG. 1) of linseed flax (*Linum usitatissimum* L.) in the experimental plots at the University. The pycnidia were separate, but inclined to be arranged in longitudinal lines, in zones 3–20 mm. in length and usually encircling the stem. This fungus appears not to have been previously reported on flax. A week later it was detected on a sample of flax from a field at Elstow, Saskatchewan. Pure cultures were readily obtained, both by culturing surface-sterilized portions of infected stems, and by spore dilutions on ordinary laboratory media. Pycnidial production was best on malt-extract agar. The fungus was again collected on April 23, 1945, in a viable condition on overwintered flax straw (FIG. 2) in the experimental plots. It was fairly common on mature plants of several varieties in the late summer.

**DISTRIBUTION.** Pre-harvest surveys in 1946 revealed that there were light infestations of the *Selenophoma* species generally distributed on flax, specimens being found at White Fox near the limit of the flax-growing area in the north of the province and extending to the Estevan district near the southern boundary. The infested area extended from the South Saskatchewan River in central Saskatchewan eastward to near the Manitoba boundary. Relatively fewer specimens were collected in the north, possibly because of the delayed maturity of the crops. The distribution of

<sup>1</sup> Contribution from the Laboratory of Plant Pathology, University of Saskatchewan, Saskatoon, with financial assistance from the Saskatchewan Agricultural Research Foundation.

<sup>2</sup> Professor of Plant Pathology.

the fungus in the western part of the province has not been determined. The wide distribution of the species of *Selenophoma* on flax as revealed by the 1946 survey suggests that the fungus was generally present before its discovery in 1944. However, an examination of a limited amount of herbarium specimens and class material collected from 1940 to 1943, and one 1923 collection, has failed to show the presence of the fungus.

**HOST RELATIONSHIPS.** In artificial inoculation of flax seed with spore suspensions or agar discs of inoculum on moist filter paper in Petri dishes there is a slight inhibition of growth in length of the roots, accompanied by curling and an increase in branch roots. No infection occurred when green flax plants in the early stages of boll development were sprayed with a spore suspension and kept in a moist chamber in the greenhouse for several days. In the field, the pycnidia are found most commonly on early maturing varieties such as Redwing; on plants affected with die-back (9), a physiological trouble in which the upper third of the plant dries up prematurely; and on plants prematurely killed from other causes, such as late root rot (a disease complex) and scorching caused by drought and hot dry winds late in the season. Occasionally, pycnidia are found on pedicels the bolls of which have failed to develop. It seems more probable that the *Selenophoma* has developed mainly as a saprophyte and that the flowers or young bolls have been blighted from some other cause; however, the possibility that under optimum conditions the fungus may show definite parasitic ability should not be ruled out. Comparison might be made with certain serious flax pathogens such as *Sphaerella linorum* Wr. (pasma) and *Polyspora lini* Laff. (browning and stem-break) which frequently do not develop until quite late in the season under the semi-arid prairie environment. Seed yield in the linseed varieties is not appreciably affected under such circumstances. At present *Selenophoma* is of no economic importance on flax. It has been found on the following linseed varieties: Arrow, Bison, Bolley's Golden, Buda, Crystal, Custer, Dakota, Malabrigo, Redson, Redwing, Rocket, Royal, Sheyenne and Victory; and on the fibre variety Stormont Cirrus. It has not been detected on wild flax (*Linum Lewisii* Pursh).



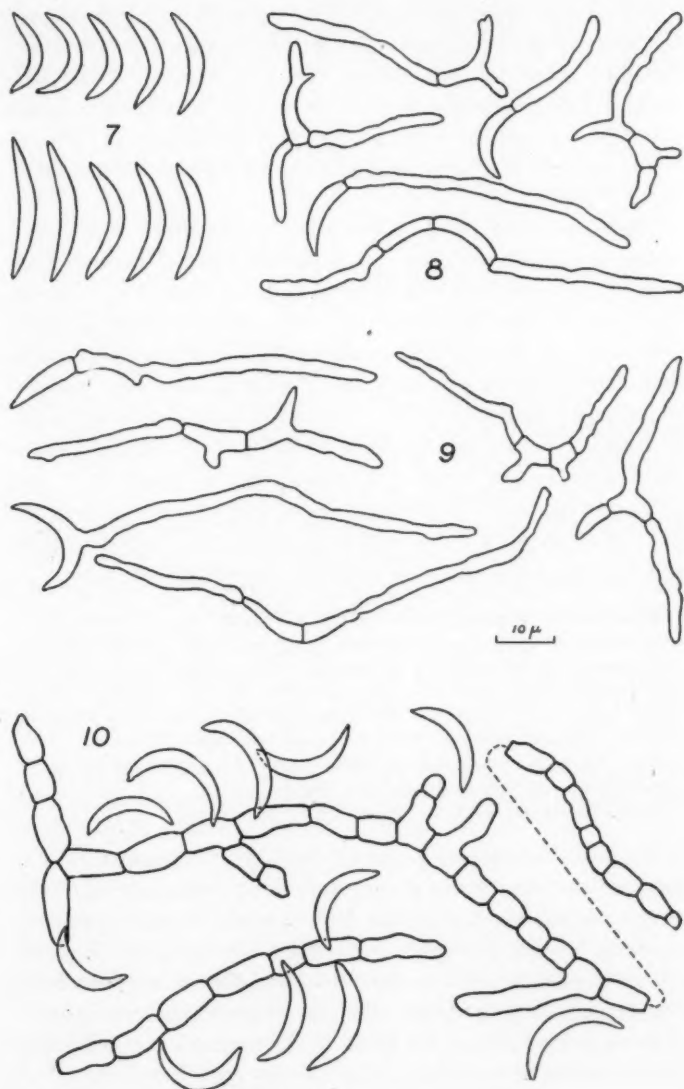
FIGS. 1-6. *Selenophoma linicola*.

During the last five years several hundred flax-seed samples have been examined for seed-borne fungi by the plating method, and no colony of *Selenophoma* has been detected. Its extremely slow growth, if it were present, and the blanketing action of the high percentage of *Alternaria* species, mostly saprophytic and commonly associated with flax seed, may possibly account for this.

It is not known how the fungus persists during the summer. As stated below, the flax form resembles *S. bromigena* (Sacc.) Sprague and Johnson (3 [*Septoria bromigena* Sacc.] and 8) more closely than any of the other species of *Selenophoma* found on the prairies (6 and 8). According to Allison (2), infections of *S. bromigena* on *Bromus inermis* Leyss. arise early in the season; this suggests that a careful search might possibly reveal *Selenophoma* leaf lesions on flax.

**MORPHOLOGY AND TAXONOMY.** Specimens of the fungus on flax were sent to Dr. Roderick Sprague, who confirmed the *Selenophoma* diagnosis and further suggested that it should be described as a new species.

In comparison with species of *Selenophoma* reported from the Northern Prairies (3, 6, and 8), spore measurements of the flax form ( $17-22 \times 2.7-3 \mu$ ) were found to be intermediate between those of *S. bromigena* ( $21-27 \times 2.8-3.5 \mu$ ), and *S. donacis* (Pass.) Sprague & Johnson var. *stomaticola* (Bauml.) Sprague & Johnson ( $13-23 \times 1.5-3 \mu$ ). In general cultural characteristics, in proneness to sectoring and in spore size and shape, the flax form is more nearly like *S. bromigena* than *S. donacis* var. *stomaticola*, though the spore measurements of the flax form from both cultural and natural material are consistently smaller than those of *S. bromigena*. Since *S. bromigena* is apparently confined to species of *Bromus* (2 and 8), cross-inoculation studies on flax and grass hosts should provide some supporting evidence on specific identity. In preliminary cross-inoculations in the greenhouse and in the field no infection was obtained on *Agropyron cristatum* (L.) Gaertn., *Agropyron pauciflorum* (Schwein.) Hitchc., *Bromus inermis*, and *Stipa comata* Trin. & Rupr., when these were inoculated with the flax form, neither did flax become infected when inoculated with *S. bromigena*.



FIGS. 7-10. *Selenophoma linicola*.

Allison (2) found spores of *S. bromigena* to be multinucleate, but those of the flax form may be either uninucleate or multinucleate. They are also usually vacuolate (FIG. 5). The terminal type of germination in water or on plain agar and the septation type on nutrient media as reported by Darley (4) for *S. bromigena* have not been found to be as clear-cut in the flax form (FIGS. 8 and 9).

The phenomenon of spore formation was observed on the mycelium on eight-day old cultures on plain and potato-dextrose agar plates prior to the formation of pycnidia (FIGS. 6 and 10). Spores were usually more numerous than those shown in these illustrations, and on the potato-dextrose agar plate the mycelium was dark and the septations closer, the individual cells being thicker-walled and chlamydospore-like. Pycnidial initials were beginning to form. *S. bromigena* behaved similarly but, in addition, occasional large, usually septate allantoid spores were observed.

On the basis of differences in spore size and host range, the flax *Selenophoma* is considered as a hitherto undescribed species and has been assigned the binomial *Selenophoma linicola*.

***Selenophoma linicola*** sp. nov. Pycnidia dispersa, saepe in rectis lineis, in maculis 3-20 mm. longis et plerumque circum caulem complexis, subepidermalia, depresso globosa, globosa vel oblonga, desuper intuentibus, brunnea vel atra, nonnihil glabra, media  $131 \times 92 \mu$ , plerumque  $110-140 \times 85-110 \mu$ ; pycnosporae unicellulares, hyalinae, vacuolatae, lunatae vel falcatae, plerumque  $17-22 \times 2.7-3.0 \mu$ , minimae  $13 \times 2 \mu$ , maximae  $26 \times 3.2 \mu$ .

Hab. In aridis, tenuibus ramis pediculisque *Lini usitatissimi* in mediis orientalibusque partibus Saskatchewan, Canada.

**Type:** Saskatoon, August, 1946, on the Royal variety.

The flax *Selenophoma* is easily cultured on the common laboratory media. The colonies are generally flat, non-lustrous, of mixed shades of dull white, gray and buff, grading through olivaceous to black, broadly concentric, with dull-white margins. Mycelial growth is slightly aerial at the center and the surface frequently develops into irregular folds. Both monosporous and multisporous cultures of most strains are given to the production of pale buff, sterile, sectors or variants (cf. 1). Radial growth on potato-dextrose and malt-extract agar is about 1 mm. in 24 hours at 22° C. Pycnidia form after about 14 days.

The pycnidia (FIGS. 1, 2, 3, and 4) are scattered, but frequently in longitudinal lines, in zones 3–20 mm. long and usually encircling the fine branches and pedicels of flax, dark brown to black, sub-epidermal but raised, flattened-globose with a slight central depression, subspherical to oblong in surface view, have thick dark peridial walls, average  $131 \times 92 \mu$  (the majority falling within a range of  $110\text{--}140 \times 85\text{--}110 \mu$ ). The ostiole measures about  $11 \times 23 \mu$  and expands up to twice these dimensions on swelling in water after spore discharge (FIG. 4).

The pycnospores (FIGS. 5 and 7) are unicellular, hyaline, lunate to falcate, uninucleate or multinucleate, usually vacuolate, typically  $17\text{--}22 \times 2.7\text{--}3 \mu$  from natural material, with a range of  $13\text{--}26 \times 2\text{--}3.2 \mu$ ; those produced in culture are larger; germination is usually preceded by septation, especially on nutrient media.

The fungus is found on dried fine branches and pedicels of linseed flax (*Linum usitatissimum* L.) at harvest time, and is distributed over the eastern half of the province of Saskatchewan. Type specimens have been placed in the Mycological Herbaria of the University of Saskatchewan, Saskatoon, and of the Division of Botany and Plant Pathology, Central Experimental Farm, Ottawa.

**SUMMARY.** A new fungus belonging to the genus *Selenophoma* is reported on dead branches and pedicels of flax in central and eastern Saskatchewan. It is the first report of a *Selenophoma* on *Linum* and is described under the binomial *S. linicola*. The fungus overwinters on flax stubble, but there is no evidence that it is seed-borne. Cross-inoculations of the flax form on certain grasses and of *S. bromigena* (Sacc.) Sprague & Johnson on flax were negative. Flax is the only known host.

**ACKNOWLEDGMENTS.** My thanks are due to Dr. Roderick Sprague, formerly of the Northern Great Plains Field Station, Mandan, North Dakota, for his willing help in determining the identity of the species and for supplying mycological specimens for comparative studies; and to Professor R. M. Ferguson, University of Saskatchewan, for his kindness in writing the Latin diagnosis.

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## DESCRIPTION OF FIGURES

FIG. 1. Pycnidia of *Selenophoma linicola* on a fine branch of a mature flax plant,  $\times 8$ . FIG. 2. Over-wintered pycnidia,  $\times 8$ . FIG. 3. Pycnidia freshly mounted,  $\times 150$ . FIG. 4. Pycnidium, after discharging spores,  $\times 200$ . FIG. 5. Spores stained with lacto-phenol-nigrosin,  $\times 900$ . FIG. 6. An eight-day culture on plain agar, showing spores produced on mycelium before pycnidia formed,  $\times 150$ .

FIG. 7. Spores of *Selenophoma linicola*. FIG. 8. Spores germinating on plain agar after fifteen hours. FIG. 9. Spores germinating on potato-dextrose agar after fifteen hours. FIG. 10. Spores produced directly on the mycelium on an eight-day plain-agar culture before pycnidia formed,  $\times 800$ .

## AN UNDESCRIBED SPECIES OF SPOROTRICHUM ON AGROPYRON<sup>1</sup>

RODERICK SPRAGUE<sup>2</sup>

(WITH 1 FIGURE)

A newly recognized parasitic species of *Sporotrichum* is widespread in the Columbia Basin of Washington and Idaho on *Agropyron spicatum* (Pursh) Scribn. & Smith, and on *A. inerme* (Scribn. & Smith) Rydb. George W. Fischer, who collected all of the material seen by the writer, found the fungus as far east as the vicinity of Bozeman, Montana, on *A. spicatum* and on *A. subsecundum* (Lk.) Hitchc. near the Yellowstone National Park at Trude, Idaho. The designated type on *A. spicatum* was collected at Roosevelt, Washington, on the Columbia River.

The lesions on the leaves are obscure, straw-colored or eventually tawny. They are scarcely distinguishable from drought-killed leaves. The infected leaves tend to become curved or recurved and show small wrinkles or blisters in the infected parts. The stromata are readily visible under the hand lens as vein-delimited, serried, nearly white stippling which resembles machine-stitching. These stromata are firm and not cottony as in *S. peribebuyense* Speg. and are  $40-150 \times 15-24 \mu$  in diameter. The hyaline mycelium is compacted, sometimes crushed, but when distinguishable the hyphae are  $2-5 \mu$  in diameter, branched and septate. It is somewhat finer than that of *S. peribebuyense* on *Setaria lutescens* (Weigel) F. T. Hubb from Boynton, Oklahoma.

<sup>1</sup> Coöperative investigations between the Divisions of Cereal Crops and Diseases, Forage Crops and Diseases, Soils, Fertilizers, and Irrigation, Bureau of Plant Industry, Soils, and Agricultural Engineering, Agricultural Research Administration; Nursery Division, Soil Conservation Service, U. S. Dept. of Agriculture; and the North Dakota and Washington Agricultural Experiment Stations.

<sup>2</sup> Formerly Pathologist, Division of Cereal Crops and Diseases, and Collaborator, Division of Forage Crops and Diseases, Northern Great Plains Field Station, Mandan, North Dakota.

There are microspores,  $2.4.5 \times 0.8-1.5 \mu$ , and macrospores,  $3-8 \times 2.5-6 \mu$ , mingled with the hyphae. The macrospores produce secondary conidia by budding and because of their yeast-like appearance together with the fact that no viable material has been available for isolation in pure culture, there remains some doubt as to whether the macrospores are part of the life cycle (FIG. 1, A). The spores of *S. peribebuyense* in the Oklahoma material, it should be added, are larger, globose, hyaline,  $8-11.2 \mu$  (FIG. 1, B), but somewhat smaller in the type. The western species is therefore distinct from the only comparable species. It in no way resembles *Fusarium Poae* (Pk.) Wr., which for some years was placed in the genus *Sporotrichum*. The new fungus shows little relation to the well known parasite of man, *S. Schenckii* (Hektoen and Perkins) Matruchet. On the basis of available information the undescribed fungus is placed in the genus *Sporotrichum* and described as new as follows:



FIG. 1: A, spore forms associated with stroma of *Sporotrichum columbiense* on *Agropyron inerme*, Newman Lake, Wash., B.P.I. 81,171. B, conidia of *Sporotrichum peribebuyense* Speg. on *Setaria lutescens*, Boynton, Okla., J. H. McLaughlin and W. W. Ray coll. Both  $\times 1000$ .

***Sporotrichum columbiense* sp. nov.** Maculis diffusis, stramineis v. fulvellis; stromatibus firmis, albidis, linearibus, in lineis dispositis,  $40-150 \times 15-24 \mu$ ; myceliis minutis v.  $5 \mu$  diam., compactis, prostratis; conidiophoris ramosis, prostratis,  $3-5 \mu$  diam.; conidiis variabilibus, subglobosis v. subcylindraceis, microsporis  $2.4.5 \times 0.8-1.5 \mu$ ; ? macrosporis  $3-8 \times 2.5-6 \mu$ .

Hab. in foliis vivis *Agropyri inermis* prope Newman Lake, Wash., June 23, 1945, B.P.I. 81,171; *ibid.*, Washtucna, Wash., June 18, 1943, B.P.I. 81,174; *Agropyri spicati*, prope Roosevelt, Wash., June 23, 1943, B.P.I. 80,934 (*typus*); *ibid.*, Belgrade, Mont., July 30, 1945, B.P.I. 81,172; *ibid.*, Whitehall, Mont., Aug. 2, 1945, B.P.I. 81,173; *ibid.*, Livingston, Mont., July 31, 1945, B.P.I. 81,175; *ibid.*, 30 miles n. of Bozeman, Mont., July 31, 1945, B.P.I. 81,176 et 81,177; *Agropyri subsecundi*, Trude, Idaho, July 28, 1945, B.P.I. 81,178. Leg. George W. Fischer.

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## SPECIES OF SYNCHYTRIUM IN LOUISIANA. IV. TWO NEW SPECIES OF SYNCHYTRIUM

MELVILLE T. COOK

(WITH 4 FIGURES)

### *Synchytrium oxalidis* sp. nov.

Gallis singulis, quarum paucae sunt in foliis petiolisque sitae,  $160\mu$  diametro, colore a flavo in aurantiacum variantibus. Gallis compositis in basibus petiolorum, magnis et soros multos continentibus. Soris flavis, aurantiacis, subrubris, circa  $22-26\mu$  diametro. Sporangiis paucis, circa  $6-8\mu$  diametro. Hab. *Oxalis repens* Thunb., Baton Rouge, La., U. S. A.

Compound galls on bases of stems and petioles of the host plants cause pronounced swellings, usually on one side (FIG. 1 A), and contain many infected cells. Small galls are few, more or less spherical, usually containing a single infected cell, and occur on petioles and leaves, and are about  $160\mu$  in diameter. The infections occur in epidermal cells (FIG. 1 B to D) but are most abundant at the bases of the petioles, which become swollen due to the stimulation of cell division and growth (FIG. 1 A, E), green, becoming light green to light yellow. The infected cells are more or less spherical and enlarge rapidly. The sori are solitary, grow rapidly and fill the infected cells except for the small amount of host cell material which may persist until the formation of the sporangia and occasionally become hard (FIG. 2 A to D). Compound galls at bases of petioles are formed by overgrowths due to successive infection of newly formed cells. The host cells surrounding the infected cells divide and grow rapidly and many of them become infected. The stimulus extends much farther from the infected cells than in any other species studied by the writer, and results in the formation of compound galls. The infected cells are submerged (FIG. 1 E to G), due to the excessive growth of surrounding tissue. Infections of these newly formed cells are so numerous that the compound galls contain a large number of in-

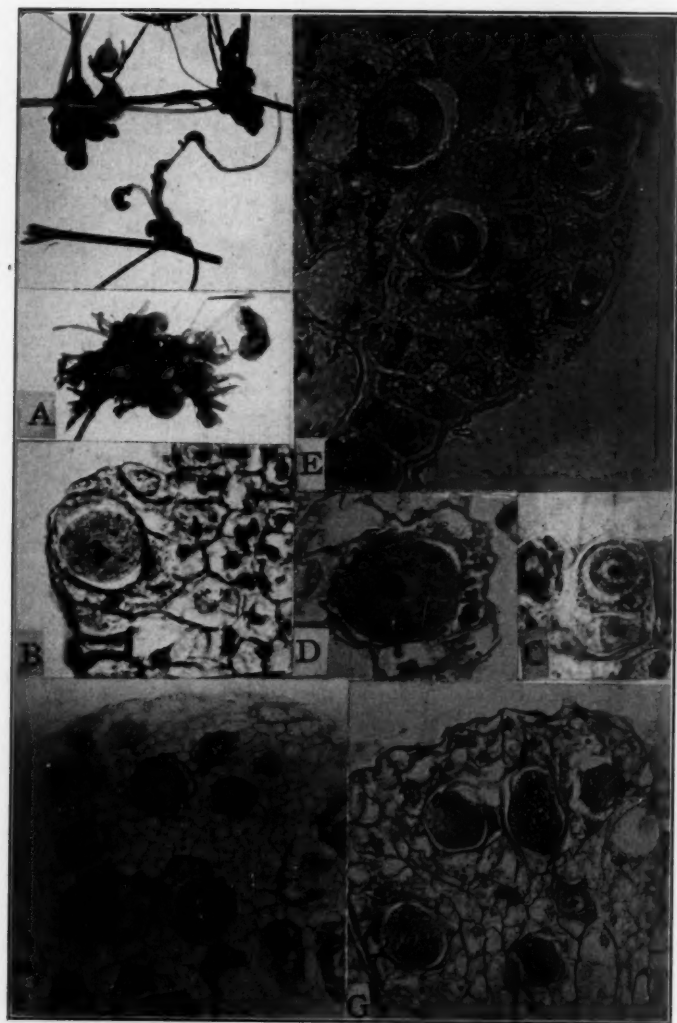


FIG. 1. *Synchytrium oxalidis*.

fect cells; the older and larger of these are submerged to a greater depth than the younger infections (FIG. 1 E, F). In some cases the newly infected cells may grow more rapidly than the older ones

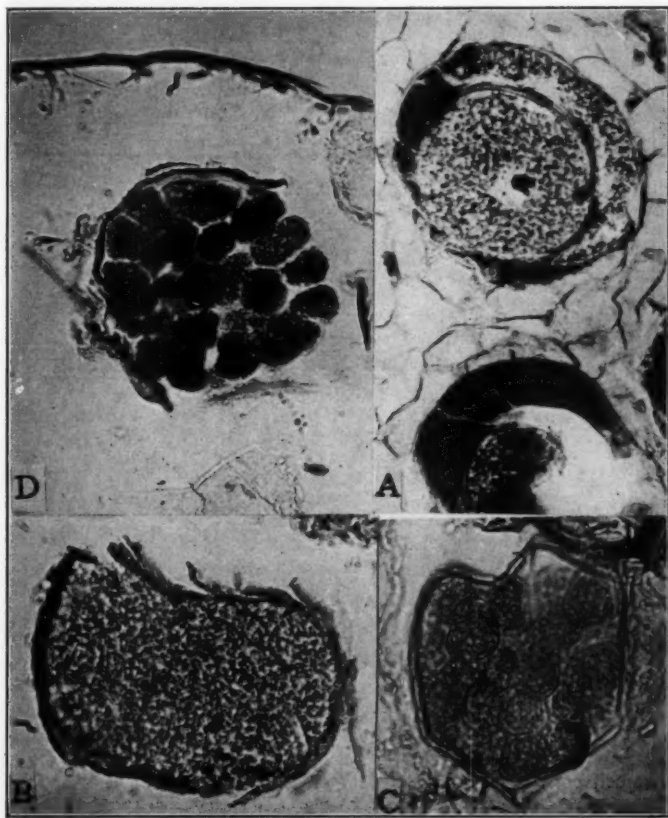


FIG. 2. *Synchytrium oxalidis*.

(FIG. 1 G). In order to get a correct idea of the relationship of old and newly infected cells to the host tissues, the section must be cut across the petiole and at right angles to the surface of the swollen region.

The sori are light green, becoming lemon yellow, orange, and finally dark red with age, and are about 22-26  $\mu$  in diameter at time

of maturity. They grow rapidly, are usually surrounded by a thick mass of host cell material which becomes granular, stains very deep and becomes hard in some cases (FIG. 1 *D, E, 2 A*). The host cell material usually decreases to form a very small zone as the time for formation of sporangia approaches (FIG. 2 *B to D*). The host cell nucleus can be seen in young infections but disappears early (FIG. 1 *C*). The nucleus of the sorus is prominent. The wall around the sorus can be seen very early, is prominent, and very thick in some cases but becomes thin with age (FIG. 1 *B to G* and FIG. 2 *A to D*). The sporangia are formed by walls uniformly throughout the sorus (FIG. 2 *B to D*). This is very similar to *S. decipiens* (Peck) Farlow. They are more or less spherical and about 6–8  $\mu$  in diameter. The host tissues degenerate early.

***Synchytrium ranunculi* sp. nov.**

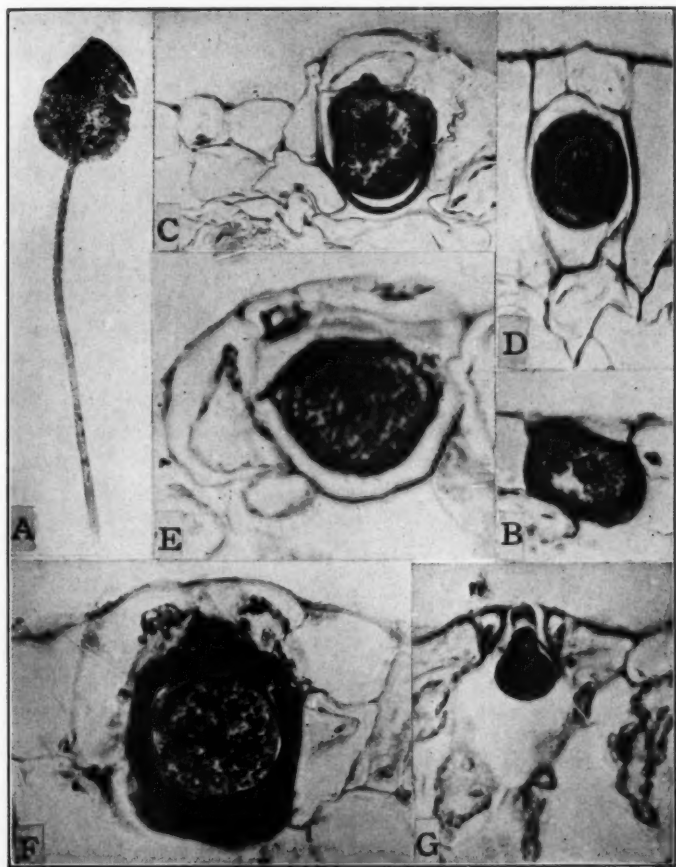
Gallis singulis, in foliis petiolisque numerosis, subflavis, subrubris nigrisque, 60  $\mu$  diametro. Soris colore a lurido in fulvum variantibus, 12  $\mu$  diametro. Sporangii 6–8  $\mu$  diametro.

Hab. *Ranunculus pusillus* Poir., Baton Rouge, La., U. S. A.

Galls single and numerous on upper surfaces of leaves and on petioles (FIG. 3 *A*), yellowish at first, become reddish and finally black, about 60  $\mu$  in diameter. The infections are in the epidermal cells (FIG. 3 *B*) which become more or less spherical, gradually enlarge and are covered with a single layer of epidermal cells (FIG. 3 *C to F*). The opening to the infected epidermal cell is very definite (FIG. 3 *F*). In a few cases, two sori were observed in the same cell (FIG. 4 *A*).

The sorus enlarges very early so as to completely fill the infected cell except for a thick layer of material which is formed from the host cell contents (FIG. 3 *B to F*). In some cases the host cell material is thick and hard (FIG. 3 *F*). The sorus is about 12  $\mu$  in diameter and is surrounded by a very definite cell wall (FIG. 3 *B to F*). The nucleus of the sorus is prominent. The fate of the nucleus of the host cell was not determined but it probably disintegrates very soon after infection. The walls of the sporangia are formed throughout the sorus as in *S. decipiens* and *S. oxalidis* (FIG. 4 *B*). The sporangia are about 8  $\mu$  in diameter and the number is much smaller than in the other *Synchytrium* reported



FIG. 3. *Synchytrium ranunculi*.

here by the author. They separate early and become more or less spherical (FIG. 4 D). In a few cases, two sori were observed in a single host cell (FIG. 4 A), but in all cases observed, only one developed.

A very unusual phenomenon was observed in this species. In some cases the sorus was observed in the substomatal cavity (FIG. 3 G). Apparently entrance was made through the stomata. In these cases the sorus was small, pear-shaped and contained a single

nucleus. The fate of these sori was not determined, but in all probability they perished for want of food since they were not in a cell.

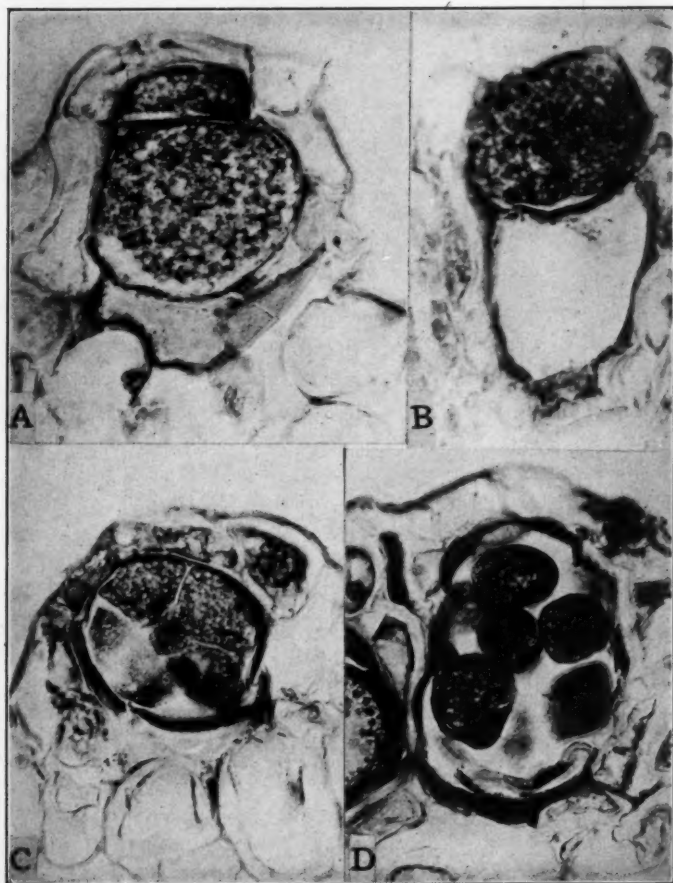


FIG. 4. *Synchytrium ranunculi*.

The writer wishes to express his thanks to Dr. C. W. Edgerton for making the photographs and to Dr. S. J. P. Chilton, Mr. Q. L. Holdeman and Mr. F. J. Reynolds for collecting the material, and

to Dr. P. G. Morehead for the Latin descriptions in this and preceding papers.

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#### EXPLANATION OF FIGURES

FIG. 1. *Synchytrium oxalidis*. A. Infected plant. B. Infected epidermal cell. C. Infected cell showing host nucleus. D. Infected cell showing host cell material. E-G. Compound galls.

FIG. 2. *Synchytrium oxalidis*. A. Infected cells showing host material surrounding sori. B. Sorus showing first indications of division to form sporangia. C-D. More advanced stages.

FIG. 3. *Synchytrium ranunculi*. A. Infected leaf and petiole. B-E. Early infections. F. Infected cell showing surrounding epidermal cells and host cell material surrounding the sorus. G. Fungus in substomatal cavity.

FIG. 4. *Synchytrium ranunculi*. A. Two sori in infected cell. B. First indication of division of sorus to form sporangia. C-D. More advanced stages.

## IMPROVEMENTS ON THE SOIL BURIAL TESTING METHOD

W. D. GRAY AND G. W. MARTIN<sup>1</sup>

(WITH 5 FIGURES)

The purposes of biological tests as applied to fabrics and other materials which are subject to deterioration brought about by the action of living organisms are: (a) to secure information which will permit a reasonable estimate of the service which may be expected in use, and (b) to indicate the comparative efficacy of substances added to the material to prevent or check the growth of organisms of decay. In many instances it is impossible to predict just what conditions a given article will encounter in service, but if it is intended for use in the moist tropics or in other situations where conditions for decay are favorable, it seems advisable to subject the article to a severe test. No one type of test has yet been devised which can be regarded as ideal. Most of the tests which have been employed in work of this nature involve either the use of pure cultures of selected organisms known to be able to attack the type of material being tested, or else a mixture of organisms, some of which are known to have the same capacity.

The advantages claimed for the pure culture methods are that greater precision is attainable with the techniques used and that there is greater assurance that tests performed in different laboratories, using the same organism and the same substratum and technique, may be correlated with each other more exactly than when other methods are used. The difficulties are the limitations of any single organism when applied to a wide range of material to be tested, the lack of reciprocal effects caused by the presence of other organisms, and the necessity for sterilization.

Mixed-culture tests may involve a mixture of known organisms, grown in pure culture and added to each other in controlled quan-

<sup>1</sup> Formerly of Jeffersonville Quartermaster Depot. Publication authorized by Office of Quartermaster General.

tity, but more commonly such tests employ a suspension of organisms secured from soil or else the material under test is buried directly in the soil. The papers cited in the bibliography describe these methods and give a number of references to earlier work which need not be repeated here. The present discussion is concerned only with soil burial, and the work of which this is a partial report was done in the Biological Laboratory of the Jeffersonville Quartermaster Depot in 1944 and 1945. Bertolet (2) had previously developed this method and found it extremely useful; his procedures are fully described in the paper cited. When the Biological Laboratory was established, one of its functions was to make a comparative study of various testing methods. A great many methods were tried and it was found that for a very wide range of material, especially textiles, the soil burial method gave the most satisfactory results. However, it was felt that this method could be improved in such a way as to remove or at least minimize the faults which had been attributed to it. These were chiefly two: the seasonal effect when burial was out of doors or even in a greenhouse, and the very great variation in the effect of different soils (5). Both of these faults contributed to the great variation reported by different laboratories when the same type of material was tested by soil burial. Another objection that has been raised against this method is the lack of light, which is reputed to exert a direct effect either on the material under test, *e.g.*, cellulose, or may produce chemical alteration in inhibitors or other materials used in finishing. Wagner, Webber and Siu (6) have shown that exposure to ultra-violet light weakens cotton fibers but at the same time renders them less susceptible to attack by *Metarrhizium glutinosum*. To what extent the pigmentation present in the finish of most articles of equipment likely to be exposed to sunlight would protect them against the effects of light has not been sufficiently investigated. Preliminary exposure to ultra-violet radiation might well be made a part of the soil burial procedure in dealing with certain types of material, but that was not done in the experiments which form the basis for this report. The criticism has been made that the soil burial test is unduly severe; however, this criticism is scarcely valid, since any test which must accomplish

in a few days what would require months in service must be correspondingly severe.

VARIATION IN RESULTS OBTAINED FROM GREENHOUSE  
BURIAL EXPERIMENTS

In greenhouse burial at Jeffersonville, an attempt was made to keep the temperature constant and the soil uniformly moist, since these two factors seem to be of prime importance. Even with experienced workers, however, this was only approximately successful, and the data presented in Table I and fig. 1 demonstrate the results of the variations in environmental factors.

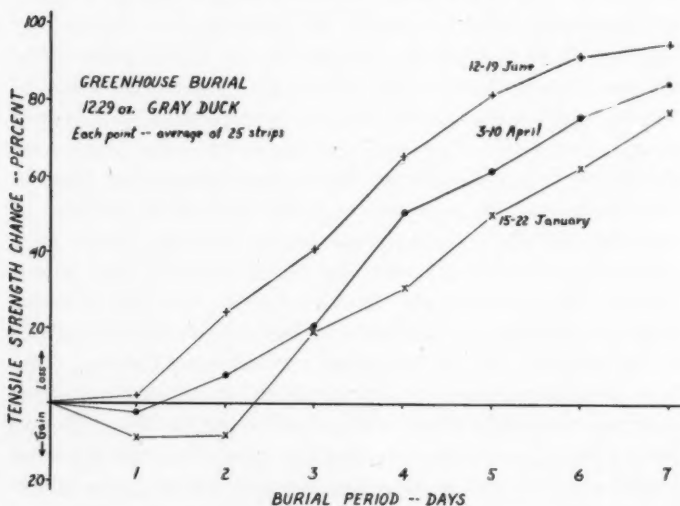


FIG. 1. Seasonal variation in greenhouse burial.

The data in Table I show that with 12.29 oz. gray duck, over seven days were required in January to reduce the breaking strength of standard strips by 80 per cent; six days were required in April and only five days were required in June to cause a comparable reduction in breaking strength. The variations reported here are in accordance with similar experiences encountered elsewhere in greenhouse burial work, and in out-of-door burial such

seasonal differences are naturally very greatly intensified. For this reason it was decided to bury test strips in soil under rather closely controlled conditions.

TABLE I  
RATE OF LOSS OF TENSILE STRENGTH OF 12.29 OZ. GRAY DUCK  
DURING GREENHOUSE BURIAL AT DIFFERENT SEASONS \*

Days Buried	January 15-22		April 3-10		June 12-19	
	Average Tensile Strength (lbs.)	Per cent Change in Tensile Strength	Average Tensile Strength (lbs.)	Per cent Change in Tensile Strength	Average Tensile Strength (lbs.)	Per cent Change in Tensile Strength
1	177.5	+8.7	178.4	+2.1	166.1	-2.1
2	176.7	+8.3	161.6	-7.6	139.3	-23.8
3	131.8	-19.2	140.2	-19.8	101.6	-40.1
4	104.9	-35.1	95.5	-45.4	57.9	-65.2
5	73.6	-54.9	68.1	-61.1	32.8	-80.6
6	56.6	-64.7	44.4	-74.6	15.5	-90.8
7	33.0	-79.8	28.8	-83.5	11.4	-93.2
Controls	163.2	—	174.8	—	169.7	—

\* In Table I and all subsequent tables, unless otherwise stated, each tensile strength figure listed is the average tensile strength of twenty-five test strips.

#### THE SOIL-PAN BURIAL METHOD

For this method a chamber in which the temperature and relative humidity could be controlled was employed. In all of the work herein reported, the temperature was maintained at  $85^{\circ}\text{F.} \pm 2^{\circ}$  and the relative humidity was maintained at  $85\% \pm 2\%$ . Test strips were buried in stainless steel soil pans which were rectangular in shape with sloping ends, 4 inches deep,  $7 \times 10$  inches on the bottom,  $7 \times 11$  inches at the top, with the upper edges ground level to permit close fitting of the glass plates,  $9 \times 13$  inches in size, used as covers. The test strips used were cut to  $1.5 \times 5.5$  inches, the long dimension being the warp, and were carefully ravelled to a width of one inch. Where FWWR duck (fire, weather, water resistant) or other heavily impregnated fabric was used, the strips were cut exactly one inch in width, care being taken not to cut a warp thread. Five or six such strips were buried in each pan, the pans were then covered with the glass plates and stacked on shelves in the controlled temperature—controlled humidity chamber.



The soil used was fresh compost, made of two parts fertile loam and one part well-rotted cow manure, the whole being carefully mixed and screened. When first used, this compost was rather slow in causing decay, but after four lots of cotton fabric had been buried in it for as many weeks, it attained maximum efficiency and thereafter maintained itself with remarkable constancy, so that the same soil was used over and over.

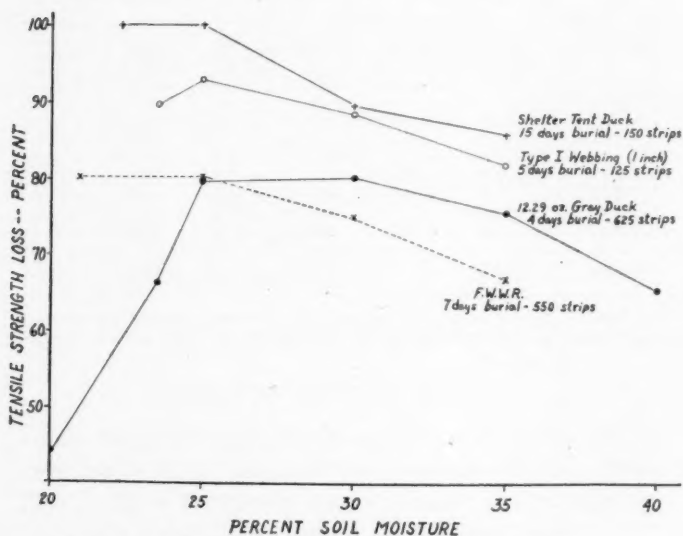


FIG. 2. Relation of soil moisture to decay of different fabrics.

It was suspected that soil moisture content might be an important factor and at first it was thought that the loose-fitting covers of the soil pans might permit the escape of water from the soil. However, preliminary experiments showed that loss of moisture within the ranges studied was not significant for periods of at least five or six weeks. The actual loss of water in five weeks from ten pans selected at random and containing soil, the initial moisture content of which was 25 per cent, varied from 0.87 to 2.28 per cent of the total moisture present. With moisture losses of such low magnitude it was possible to run experiments for a period of several weeks and maintain the soil moisture percentage nearly constant

at whatever value it had initially been set. Results of breaking-strength measurements of nearly fifteen hundred test strips which had been buried in soil of varying moisture contents demonstrated that variations in soil moisture induced great variations in rate of loss of tensile strength. Figure 2 shows the relationship of soil moisture content to tensile strength loss of four different fabrics. It should be noted that a soil moisture content of 25 per cent gave

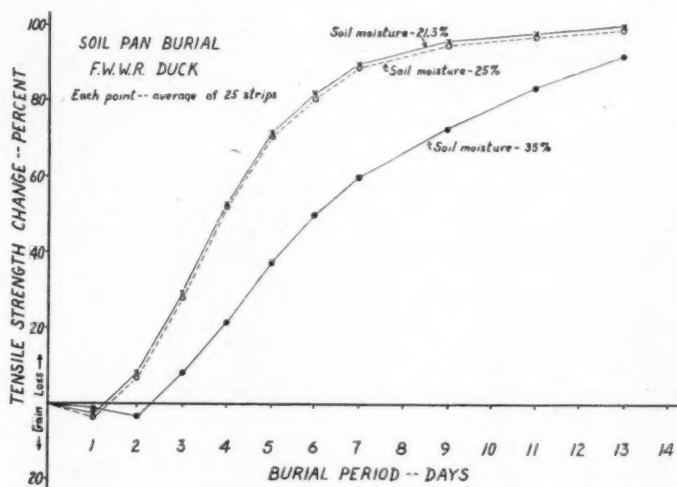


FIG. 3. Effect of varying moisture content on decay of F. W. W. R. duck.

a maximum rate of decay for three of the four fabrics tested; only in the case of the heavy gray duck was the rate very slightly, but not significantly higher at 30 per cent moisture content. In the case of shelter-tent duck there was little difference in rate of decay over a moisture range of 22-25 per cent, and with F.W.W.R. duck the rate was about the same over the moisture range of 21-25 per cent. Figure 3 shows the very close approximation between the tensile strength losses of F.W.W.R. duck at 21.3 per cent and 25 per cent soil moisture as compared with the sharp drop in rate of decay when the moisture content was raised to 35 per cent. On the basis of these and other experiments a soil moisture content of 25 per cent, based on the dry weight, was adopted as the standard.

For soils of similar texture, this moisture content should result in nearly maximum rate of decay; however, for soils with dissimilar water-holding capacities, it is quite probable that another moisture value would prove optimum.

Certain irregularities appeared from time to time as may be seen in fig. 4. Shelter-tent duck treated with water repellent and buried in soil of 25 per cent moisture content decayed twice as fast as the

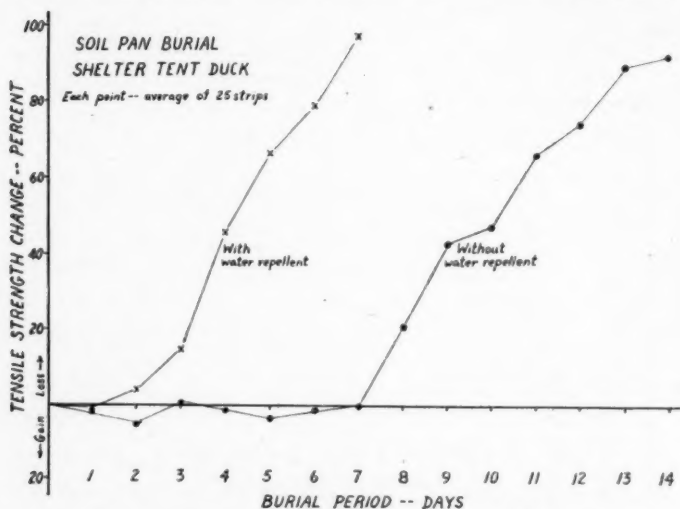


FIG. 4. Effect of repellent on decay of shelter tent duck.

same fabric without the water repellent. The nature of the water repellent treatment was unknown; however, it may be that the material used in the treatment actually contained some nutrient substances which were available to the fungi present and acted as growth stimulants. On the other hand, this phenomenon might possibly be explained on purely physical grounds, since the material which contained no water repellent became quite wet when buried in soil and may have been too wet for rapid growth of the fungi involved. Such irregularities appear in any type of test and require individual interpretation, and, where possible and important enough to justify the expenditure of time, comparison with the results of other types of test on the same material.

Of more general significance is the effect of leaching. Figure 5 and Table II present the results obtained when F.W.W.R. duck containing 0.35 per cent copper (half as copper naphthenate and half as basic copper oleate) was buried in soil pans and in the greenhouse, with and without preliminary leaching for 72 hours.

TABLE II

COMPARATIVE RATES OF BREAKDOWN OF LEACHED AND UNLEACHED F.W.W.R. DUCK CONTAINING 0.35 PER CENT COPPER (HALF AS COPPER NAPHTHENATE AND HALF AS BASIC COPPER OLEATE) DURING SOIL PAN BURIAL AND GREENHOUSE BURIAL

Days Burial	Percentage Change in Tensile Strength			
	Soil Pans		Greenhouse	
	Unleached	Leached	Unleached	Leached
4	+5.1	-1.9	+2.0	+1.2
8	+4.9	-4.2	+1.6	-3.0
12	+4.8	-49.3	-0.3	-6.3
16	+1.4	-86.4	-18.5	-11.2
20	-2.3	-91.8	-22.4	-22.2
24	+3.1	-99.3	-47.9	-44.9
28	-9.5		-62.8	-52.7
32	-22.3		-72.1	-66.9
36	-35.2		-81.5	-75.6
40	-68.3		-86.8	—
44	-76.9		-94.9	-83.4
48	-87.4		-86.2	
52	-100.0		-68.0	
56	-100.0		-74.8	
60	-100.0		-94.8	

It will be noted that there is little significant difference between the unleached and leached strips in greenhouse burial, whereas in the soil pans the leached fabric was completely destroyed in 24 days while the unleached material did not show a 100 per cent loss in tensile strength until 52 days had elapsed. These results bring out an important difference between greenhouse burial, where the frequent watering provides a leaching effect, and the soil pans, where the limited space and relatively constant soil moisture tend to hold inhibitors in the fabric or in the immediately adjacent soil. It is obvious that when inhibitors are present or are suspected of being present, material to be tested in soil pans should be leached before burial, since fastness to leaching is an important characteristic of an inhibitor. A large number of tests has established a three-day

leaching period as ample for practically all types of inhibitor. Even with the added time necessary for such preliminary treatment, the soil-pan burial method is much more rapid in its results than greenhouse burial. In the instance illustrated, which is fairly repre-

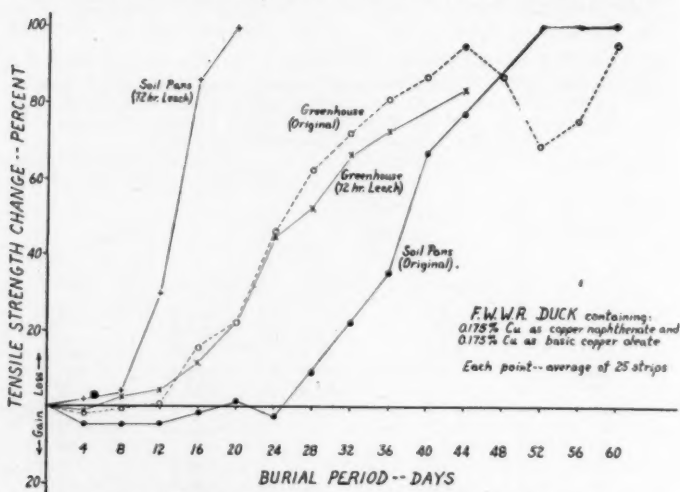


FIG. 5. Effect of leaching on decay of F. W. W. R. duck.

sentative of many other tests, deterioration was approximately twice as rapid in the soil pans as in the greenhouse. Attention may also be called to the much greater regularity of the curves derived from the results of soil-pan burial.

#### CONCLUSION

With properly prepared and biologically active soil in standard containers where the soil moisture content can be accurately regulated and maintained, and incubation under conditions of closely controlled temperature and relative humidity, soil burial tests of material subject to biological deterioration can be conducted with a high degree of accuracy and reproducibility. Under such conditions, soil burial is a rapid test of great significance, especially when the materials being tested are fabrics.

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## A SIMPLE AND RAPID METHOD FOR OBTAINING MONOSPORE CULTURES OF FUNGI

LUCILLE K. GEORG

(WITH 2 FIGURES)

The isolation of spores  $2-3\ \mu$  or larger may be accomplished readily by the use of a small glass conical tip which is attached to the low power objective of the microscope. The larger opening of the cone fits around the lens of the objective (see FIG. 1),

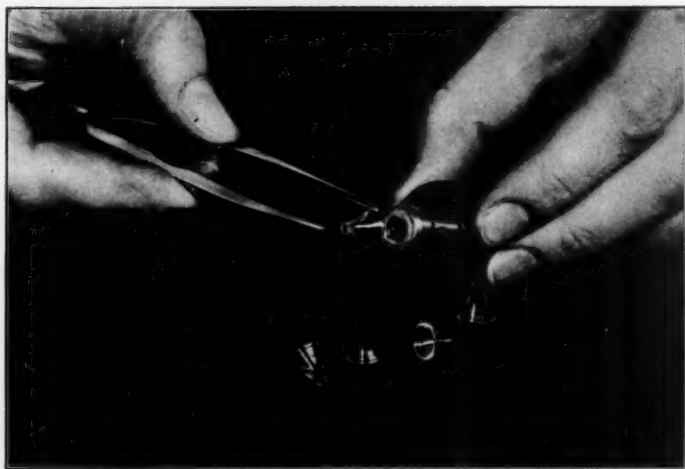


FIG. 1. Glass tips for isolating spores.

and the smaller opening approximates the microscopic field as seen through that lens.

Several such conical tips may be drawn out from glass tubing, preferably pyrex. To make the conical tip, glass tubing is selected which has an inside diameter just slightly larger than the diameter of the lens of the low power objective to be used. A small area



of the tubing is heated to redness by rotating it in a very hot flame and is then very quickly drawn out so that a steep cone is produced between the original tubing and the drawn out end. This portion can be separated by using a sharp file and breaking at the tip and base of the cone. The following criteria are essential in preparing an accurate tip:

1. The inside diameter of the tubing (which will be the inside diameter of the large end of the finished cone) must be slightly larger than the diameter of the lens of the low power objective so the larger end of the cone will just fit around this lens (approximately 6 to 7 mm. inside diameter).

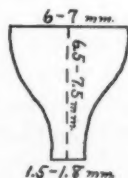


FIG. 2. Measurements of glass tip.

2. The smaller end of the conical tip should have a diameter which approximates that of the microscopic field as seen through the low power objective. For an objective of  $8\times$  magnification used with a  $10\times$  ocular, this field is approximately 1.7 mm. in diameter.

3. The length of the conical tip must be slightly less than the working distance of the objective—*i.e.*, less than the distance between the front lens of the objective and the object on which it is focused. For the  $8\times$  objective used, the working distance is approximately 8 mm. and the conical tips average between 6.5 to 7.5 mm. long (FIG. 2).

#### METHOD OF SPORE ISOLATION

I. Preparation of a dilute suspension of spores: A dilute suspension of spores is obtained by pouring sterile water onto a well developed culture of the fungus, preferably on a slant of solid medium, and then rotating the tube between the hands or gently shaking for several minutes. A loop of the fluid may be examined

to determine whether spores have been washed from the culture. The amount of water to be added will depend on the number of spores that can be washed from the mycelium.

II. Seeding of cornmeal agar plates: Cornmeal agar is used because it is very clear and the spores can be seen quite easily on its surface. Also, it has proved to be a good medium for the germination of the spores studied.

A loop of dilute spore suspension is spread over the surface of a thin cornmeal agar plate. (The agar should not have a depth of more than 2 mm.) Serial plates may be inoculated with the same loop to insure that a plate will be obtained which will have well dispersed spores.

The spores are allowed to germinate at room temperature, and on the second or third day the plates may be opened and examined with the low power lens for sprouting spores. The spores must be so dispersed that areas can be found where only one spore occurs in a microscopic field.

III. Isolation of a single sprouting spore: The low power objective is removed from the microscope and a very thin ring of plasticine is fitted around the lens. A sterile conical glass tip is taken in a forceps and fitted over the lens gently pressing against the plasticine ring to which it will adhere (see FIG. 1). The objective is then returned to the microscope and focused on the surface of the agar. When a field is found where only one spore is present, the spore is centered in the microscopic field, and the objective is carefully lowered so that the small end of the conical tip will cut through the agar and form a ring around the spore. The objective is raised and the small disc of agar within the cut circle must be carefully examined to determine that only one spore is present in this area. The area just outside the circle should also be examined so that there is no danger of contamination by another spore which may lie just outside. The disc of agar is then removed from the plate using a fine flattened sterile needle and is placed right side up on a fresh plate of medium of a type which will support good growth of the organism under study. This plate may then be examined in order to determine that the spore has actually been transferred on the small disc of agar. The

plate may be further examined on successive days to make sure that only one spore is growing on the plate.

The glass tip may be resterilized by removing it from the objective and holding it in the flame for a few seconds. When it cools it may be reattached and another spore isolated.

This method has proved to be successful for obtaining monospore cultures of *Trichophyton*.

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## HAPLOSPORANGIUM IN CANADIAN RODENTS<sup>1</sup>

ELEANOR SILVER DOWDING<sup>2</sup>

In 1942 Emmons (1) described *Haplosporangium parvum* which he isolated from the lungs of wild Arizona rodents. *H. parvum* is a Phycomycete which on artificial media grows as a septate mycelium bearing single-spored sporangia, and in the lung appears as a large thick-walled oval or spherical cell (chlamydo-spore).

During the summer of 1946, collections of the lungs of wild rodents were made in Western Canada<sup>3</sup> over an area of about 1000 square miles extending from Lethbridge to Peace River. Two hundred and seventy-five animals (nine species) were collected, one portion of the lung of each animal being preserved and another portion being planted in Sabouraud's medium.

It was found that fourteen animals possessed fungous cells in their lungs. The hosts were thirteen white-footed deer-mice (*Peromyscus maniculatus borealis*) and one red squirrel (*Sciurus hudsonicus baileyi*). From the lungs of eight of these infected animals *Haplosporangium parvum* was obtained in culture. The range of *Haplosporangium parvum* has therefore been extended into Western Canada, and two new hosts have been reported—the common white-footed mouse, and the red squirrel.

In Arizona the largest *Haplosporangium parvum* cell recorded within the lung was 45  $\mu$  in diameter. In Alberta the chlamydo-spore may reach the relatively enormous dimensions of 270  $\mu$  and its wall may be 8–10  $\mu$  thick.

Fungous cells in Canadian rodent lungs have never been observed to contain endospores. Some of the cells in Arizona rodent lungs were filled with endospores (2). Since the Arizona ro-

<sup>1</sup> This investigation was made possible by financial assistance from the National Research Council of Canada.

<sup>2</sup> Mrs. E. S. Keeping.

<sup>3</sup> Collections were made by the Field Survey of the Division of Entomology of the Province of Alberta.

dents were known to be infected with *Coccidioides immitis* as well as with *Haplosporangium parvum*, and since no coccidioidal infection was encountered in our Canadian rodent survey, it would seem possible that the endosporulating cells in the Arizona material were those of *C. immitis* and not of *H. parvum*.

*Haplosporangium parvum* has been cultivated upon soil in the Provincial Laboratory. It grows vigorously upon this medium and produces sporangia. It was found that the sporangia could not be shed by jarring the culture nor could they be dispersed by air currents. They are adhesive and are readily transferred by contact. It may be inferred from these observations that the fungus naturally grows upon soil. There burrowing rodents come into contact with it, and carry away the adhesive sporangia on their faces. When the animals clean themselves with their paws, the sporangia may be inhaled.

Although the spores can readily be induced to germinate at room temperature, yet at 37° C. no germination takes place. The sporangia merely swell. After three weeks of incubation they become as much as ten times their original diameter and are then large thick-walled chlamydospores resembling those found naturally in the mouse lung, although about one-fifth the size. These chlamydospores when removed from the incubator and transferred to soil-agar germinate within two days, producing a number of radiating germ-tubes. It may be inferred from these observations that the sporangium, after it is inhaled by the mouse, swells within the lung to form a large thick-walled chlamydospore which after the death of the animal germinates in the soil and so initiates the saprophytic phase.

A detailed account of the work will be published shortly (3).

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#### LITERATURE CITED

1. Emmons, C. W., and Ashburn, L. L. Public Health Reports 57: 1715-1727. 1942.
2. Ashburn, L. L., and Emmons, C. W. Archives of Pathology 39: 3-8. 1945.
3. Dowding, E. S. Canadian Journal of Research (*in press*).

## NOTES AND BRIEF ARTICLES

### COPROPHILOUS ASCOMYCETES OF VIRGINIA

Dung was collected from pastures, open woods, barns and manure piles in the following counties: Albemarle, Augusta, Campbell, Clark, Giles, Montgomery, Norfolk, and Nottoway. Some of it was old and weathered, some several days old, and some freshly dropped. The dung was placed in pans lined with moist paper towels. Dry dung was moistened with water, and water was added to the cultures from time to time to prevent their drying out. They were kept in diffused light at room temperature. The pans were covered with glass panes to prevent rapid evaporation and also to protect the cultures. Rabbit and deer mouse dung, consisting of small balls, was placed in Petri dishes to facilitate handling.

At intervals of two to three days a portion of dung was removed from the culture pan and examined under a wide field binocular microscope. Ascocarps were lifted from the dung with dissecting needles and mounted on slides in a 10 per cent glycerine solution. Old, weathered dung from open fields produced more ascocarps than fresh dung or dung that had been packed in manure piles. It was necessary when mounting the hairy perithecia of *Chaetomium* to use first a solution of alcohol, glycerine, and water to avoid capturing air bubbles in the hairs of spore masses. After a few minutes this solution was drained off and the glycerine solution added. The specimens were then identified and catalogued.

The following thirteen genera and sixty species of coprophilous Ascomycetes have been observed.

#### DISCOMYCETES

*Ascobolus americanus* (Cook & Ellis) Seaver. On horse dung, Charlottesville, April 1, 1942. *A. geophilus* Seaver. On horse dung, Charlottesville. *A. glaber* Pers. On horse dung, Charlottesville, January 21, 1942. Also found on horse dung collected

at Boyle, Mississippi. *A. immersus* Pers. On horse dung from Charlottesville, Norfolk, and Blandy Farm (Boyce, Va.), November 12, 1941. *A. Leveillei* Boud. On horse dung from Charlottesville, January 19, 1942. *A. magnificus* Dodge. On cow dung from Charlottesville, February 9, 1942. *A. stercorarius* (Bull.) Schröt. On cow and horse dung from Blackstone, Charlottesville, Norfolk and Blandy Farm, November 12, 1941. *A. striisporus* (Ellis & Dearn.) Seaver. On horse dung, Charlottesville. *A. viridulus* Phill. & Plow. On rabbit dung from Elliott Knob, May 25, 1942. *A. Winteri* Rehm. On horse dung, Charlottesville, November 28, 1941. *Saccobolus Kerverni* (Crouan) Boud. On horse dung from Charlottesville, Norfolk, and Blackstone, November 11, 1941. *S. violascens* Boud. On horse dung, Norfolk, January 31, 1942. *Ascophanus argenteus* (Curr.) Boud. On horse dung from Charlottesville. *A. carneus* (Pers.) Boud. On cow dung from Lynchburg, April 3, 1942. On rabbit dung from Elliott Knob, May 25, 1942. *A. lacteus* (Cooke & Phill.) Sacc. On horse dung, Charlottesville, May 27, 1942. *A. vicinus* Boud. On horse dung, Charlottesville, January 19, 1942. *Ryparobius crustaceus* (Fuckel) Rehm. On horse dung, Charlottesville. *R. sexdecimsporus* (Crouan) Sacc. On horse dung, Charlottesville, May 14, 1942. *Thecotheus Pelletieri* (Crouan) Boud. On horse dung, Lynchburg, June 3, 1942. *Lasiobolus equinus* (Müll.) Karst. On horse dung, Charlottesville, March 30, 1942. *Peziza fimeti* (Fuckel) Seaver. On cow dung, Charlottesville. *P. vesiculosa* Bull. ex Fr. On cow dung, Charlottesville, January 31, 1942.

## PYRENOMYCETES

*Chaetomium ampullare* Chivers. On rabbit dung, Elliott Knob, June 5, 1942. *C. bostrychodes* Zopf. On horse dung from Norfolk, May 20, 1942. Also on deer mouse dung from Mountain Lake. *C. caprinum* Bainier. On rabbit dung, Elliott Knob, June 4, 1942. *C. crispatum* Fuckel. On rabbit and horse dung, Charlottesville, November 17, 1942. *C. globosum* Kunze. On horse dung, Norfolk, May 16, 1942. *C. murorum* Corda. On horse dung, Norfolk, May 18, 1942; Radford, May 7, 1942. *C. sub-*



*spirale* Chivers. On rabbit dung, Elliott Knob, May 27, 1942. *Sordaria fimicola* (Rob.) Ces. & DeNot. On horse and cow dung, Charlottesville. *S. humana* (Fuckel) Awd. On horse dung from Radford, April 27, 1942; also from Norfolk. *S. leucoplaca* (B. & R.) E. & E. On horse dung, Charlottesville, April 21, 1942. *S. macrospora* Awd. On horse dung, Charlottesville, January 24, 1942; Lynchburg, April 1, 1942; Blandy Experimental Farm, April 27, 1942. *S. minima* (Sacc. & Speg.) Sacc. On horse dung, Charlottesville, April 9, 1942. *Pleurage adelura* Griffiths. On horse dung Charlottesville, April 13, 1942. *P. albicans* (Alb. & Schw.). On horse and cow dung, Charlottesville. *P. amphicornis* (Ellis) Kuntze. On rabbit dung, Charlottesville, May 25, 1942. *P. anomala* Griffiths. On horse dung, Norfolk, January 13, 1942; Lynchburg, April 1, 1942. Griffiths and Seaver (1910) indicate it as known only from the type locality, New York. *P. anserina* (Rabh.) Kuntze. On horse dung, Norfolk, April 1, 1942. On horse dung, Lynchburg and Charlottesville. Also found on horse dung from Boyle, Mississippi. *P. arachnoidea* (Niessl.). On rabbit dung, Charlottesville, May 8, 1942. *P. collapsa* Griffiths. On horse dung, Charlottesville, March 14, 1942; also on rabbit dung from Elliott Knob. *P. conica* (Fuckel) Griffith & Seaver. On rabbit dung, Elliott Knob, June 5, 1942. On cow dung, Lynchburg, April 3, 1942. *P. curvicolla* (Wint.) Kuntze. On rabbit dung from Charlottesville and Elliott Knob, May 8, 1942; also on horse dung from Charlottesville. *P. dakotensis* Griffiths. On horse dung, Charlottesville, May 8, 1942. *P. decipiens* (Wint.) Kuntze. On horse dung from Charlottesville, May 8, 1942. *P. erostrata* Griffiths. On horse dung from Charlottesville, May 6, 1942, and from Norfolk. Also found on horse dung collected by Mr. C. W. Merritt from Boyle, Mississippi, January 15, 1942. *P. fimiseda* Ces. & DeNot. On horse dung from Radford, June 5, 1942. *P. minuta* (Fuckel) Kuntze. On horse dung from Blandy Experimental Farm, April 29, 1942. *P. pleiospora* (Wint.) Kuntze. On cow dung from Charlottesville. *P. taeniodes* Griffiths. On horse dung, Charlottesville, November 24, 1941; Lynchburg, March 30, 1942. *P. vestita* Zopf. On horse dung, Charlottesville, March 14, 1942; Lynchburg, April 10, 1942; Blackstone, May 1, 1942. *P. zygospora* (Speg.)

Kuntze. On horse dung, Charlottesville, November 18, 1941. *Hypocopra gigaspora* E. & E. On cow dung, Charlottesville. *Delitschia leporina* Griffiths. On horse dung, Charlottesville. *Sporormia chaetomioides* Griffiths. On horse dung, Lynchburg, May 2, 1942. *S. corynespora* Niessl. On cow dung, Charlottesville. *S. herculea* E. & E. On horse dung, Lynchburg, May 2, 1942. *S. intermedia* Awd. On horse dung, Charlottesville, April 13, 1942; Blandy Experimental Farm, April 27, 1942. *S. leporina* Niessl. On horse dung, Charlottesville, November 11, 1941. *S. minima* Awd. On horse dung, Charlottesville, November 11, 1941.

The author wishes to thank Dr. E. M. Betts, University of Virginia, under whom this paper was worked out, and also the numerous contributors of material collected all over the state.—CHARLES MAYE WILSON, University of Virginia.

#### NOTICE

The annual foray of the Mycological Society will be held at Highlands, North Carolina, Sept. 2-7. Write to Prof. J. H. Miller, Dept. of Plant Path., Univ. of Georgia, Athens, Ga., in regard to accommodations. Please give the number in your party and length of stay.—ALEXANDER H. SMITH.



